Universidade do Minho
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Clinical experimental trials on changes in ocular surface induced by soft contact lenses wear

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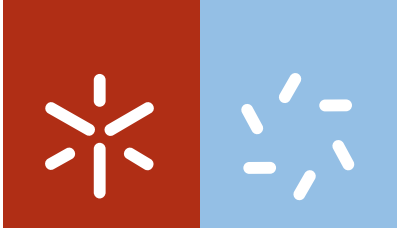

Universidade do Minho
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Kishor Sapkota

Clinical experimental trials on changes in ocular surface induced by soft contact lenses wear

Ph.D. Thesis in Sciences
Specialization in Physics

Supervisors:
Professora Doutora Maria Madalena Cunha Faria Lira
Professora Doutora Sandra Maria de Braga Franco

November 2015

STATEMENT OF INTEGRITY

I hereby declare having conducted my thesis with integrity. I confirm that I have not used plagiarism or any form of falsification of results in the process of the thesis elaboration. I further declare that I have fully acknowledged the Code of Ethical Conduct of the University of Minho.

University of Minho, 19 November 2015

Full name: Kishor Sapkota

Signature: KSapkota

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Dedicated to my parents

Khim Lal Sapkota & Harimaya Sapkota

Who stay tens of thousands of miles away and constantly pray for my progress and success.

List of publications

This thesis is based on the following original articles:

Sapkota K, Franco S, Sampaio P, Lira M. Goblet cell density association with tear function and ocular surface physiology. Contact Lens Anterior Eye, 2015;38(4):240-4.

Sapkota K, Franco S, Sampaio P, Lira M. Effects of three months of soft contact lens wear on conjunctival cytology. Clin Exp Opt 2015 in Press.

Sapkota K, Franco S, Lira M. Intraocular pressure measurement with ocular response analyzer over soft contact lens. Contact Lens Anterior Eye, 2014;37(6):415-9.

Sapkota K, Franco S, Lira M. Ocular surface physiology changes with three months of soft contact lens wear. Submitted to Optometry and Vision Science in October 2015.

Sapkota K, Franco S, Lira M. Effect of soft contact lens wear on corneal biomechanical properties. Submitted to Clin Exp Optom in October 2015.

Sapkota K, Franco S, Lira M. Effect of three months of soft contact lens wear on intraocular pressure. Submitted to Contact Lens Anterior Eye in September 2015.

Sapkota K, Franco S, Lira M. Contrast sensitivity function with soft contact lens wear. Paper in the process of submission.

Summary

Contact lenses (CL) are an important method of refractive error correction. Every year, millions of people commence CL wear. About 140 million people in the world wear CL and almost 90% of them wear soft contact lenses (SCL). Every year, several lenses with different materials, designs or chemical composition come in the market. It is becoming one of the important businesses with billions of dollars investment. As CL are placed directly on eyes, it may affect the cornea, conjunctiva, eyelids, and tears either mechanically, physiologically and/or immunologically. The present thesis work was developed to analyse some of these aspects.

The main objective of this thesis was to study the changes in ocular surface induced by SCL. Several experiments were conducted to study the effect of SCL wear on the cornea, conjunctiva and tear film in more than 75 subjects who had never worn lenses before. One of the objectives was to determine the goblet cell density (GCD) in healthy eyes of non-CL wearers and to find out the relationship between GCD and tear function and ocular surface physiology. It was also investigated the changes on conjunctival epithelial cell morphology and GCD with SCL wear. Conjunctival impression cytology was performed on superior bulbar conjunctiva for this cytological experiment. The next experiment was designed to determine the effect of SCL wear on conjunctival bulbar and limbal redness and conjunctival and corneal staining. Changes on corneal biomechanical properties [corneal resistance factor (CRF) and corneal hysteresis (CH)] with CL wear were also evaluated with the Ocular Response Analyzer (ORA). The objectives of the experiments on intraocular pressure (IOP) were to investigate the accuracy of IOP measurement over SCL and to determine the changes in IOP after three months of CL wear. It was measured Goldmann-correlated intraocular pressure (IOPg) and corneal-compensated intraocular pressure (IOPcc) with ORA. The effect of CL wear on contrast sensitivity function (CSF) was evaluated with CSV-1000 VectorVision.

From the experiments on conjunctival cytology, it was observed high variation on GCD on the superior bulbar conjunctiva and the stability of the tear film was correlated with this parameter. SCL wear affected conjunctival GCD and the reduction observed in GCD was dependent upon lens materials. Epithelial cell metaplasia grading increased by at least one grade in more than two-thirds of the eyes, but this was not statistically significant. SCL wear increased conjunctival bulbar

and limbal redness, conjunctival and corneal staining independent upon the wearing modality. Even the lenses with hyperpermeability materials could not maintain the same ocular surface physiology. It was associated with lens materials and the increment was higher during the early period of wear. Higher conjunctival redness was observed on the temporal and nasal region while higher corneal staining was observed on the inferior cornea.

It was also found that SCL wear affected corneal biomechanical properties, however, different properties were affected differently. CRF reduced significantly and was associated with lens materials, but no change was observed on CH. It was found that ORA underestimates IOP measured over SCL and this underestimation was more than 3 mmHg in about one-third of the eyes. Moreover, IOPcc was more affected than IOPg. It was observed that three-months of SCL wear reduced IOP and this was associated with lens materials as well as corneal properties. In the present study it was found that CSF was better with CL than with spectacles and three months of SCL wear did not change the CSF in any spatial frequencies.

In general, these experiments showed that a variety of changes can occur on the ocular surface by three-months of SCL wear which were associated with lens materials. Reduction in GCD affects the tear film stability which may cause lens-related dry eye. Current CL with high oxygen permeability also affect ocular surface physiology like conjunctival redness and corneal staining. Although these changes can be clinically non-significant, these can affect the comfort level of lens wearers. Effect of SCL wear on IOP should be remembered on the treatment of glaucoma risk subjects. Since the same lens material affects differently on conjunctival cytology or ocular surface physiology or on IOP, there may not be an ideal CL for every subject. Lens materials should be selected for a particular subject depending upon his/her ocular health status.

Resumo

As Lentes de Contacto (LC) são um importante método de compensação dos erros refrativos. Todos os anos, milhões de pessoas iniciam o uso de LC. Cerca de 140 milhões de pessoas no mundo usam LC e aproximadamente 90% destes usam lentes de contacto hidrófilas (LCH). Todos os anos, várias lentes com diferentes materiais, desenhos ou composições químicas entram no mercado. Está a tornar-se um dos mais importantes negócios com biliões de dólares de investimento. Uma vez que as LC são colocadas diretamente nos olhos, podem afetar a córnea, conjuntiva, pálpebras e lágrima tanto mecânica, fisiológica como imunologicamente. O presente trabalho de doutoramento foi desenvolvido de forma a avaliar alguns destes aspetos.

O principal objetivo desta tese foi estudar as alterações na superfície ocular induzidas pelo uso de LCH. Foram levados a cabo vários ensaios de forma a estudar o efeito do uso das LCH na córnea, conjuntiva e no filme lacrimal em mais de 75 indivíduos que nunca tinham usado lentes. Um dos objetivos foi determinar a densidade de células caliciformes (DCC) em olhos saudáveis em não utilizadores de LC e encontrar a relação entre a DCC, a função lacrimal e a fisiologia da superfície ocular. Foram também pesquisadas as alterações na morfologia das células epiteliais e DCC com o uso das LCH através de citologia de impressão conjuntival na conjuntiva bulbar superior. Foi ainda determinado o impacto do uso de LCH na hiperemia da conjuntiva bulbar e limbal, tingido corneal e conjuntival. As alterações nas propriedades biomecânicas da córnea [fator de resistência corneal (CRF – *Corneal Resistance Factor*) e histerese corneal (CH – *Corneal Hysteresis*)] com o uso de LC foram também avaliadas com o *Ocular Response Analyzer* (ORA). A avaliação da pressão intraocular (PIO) serviu para investigar a exatidão da medição da PIO sobre as LCH e determinar ainda as alterações neste parâmetro após três meses de uso de LC. Foi ainda medida a pressão intraocular correlacionada Goldmann (PIOg) e pressão intraocular compensada (PIOcc) com o ORA. O efeito do uso de LC na sensibilidade visual ao contraste (SVC) foi também avaliado, utilizando-se para isso o CSV-1000 VectorVision.

Nos estudos realizados para a avaliação da citologia conjuntival, observou-se uma grande variação da DCC da conjuntiva bulbar superior e encontrou-se um correlação entre a estabilidade do filme lacrimal e este parâmetro. O uso de LCH afetou a DCC conjuntival e a redução observada foi dependente do material das lentes. Na classificação da metaplasia das células epiteliais

observou-se um aumento de pelo menos um grau em mais de dois terços dos olhos, mas esta diferença não foi estatisticamente significativa. O uso de LC provocou um aumento da hiperemia conjuntival bulbar e limbal mas os tingidos corneal e conjuntival não apresentaram relação com a modalidade de uso das lentes. Mesmo as lentes com materiais hiperpermeavéis não conseguiram manter a fisiologia da superfície ocular. Estas alterações mostraram estar relacionadas com os materiais das lentes e o incremento foi maior durante o período inicial de uso. A maior hiperemia conjuntival foi observada na região temporal e nasal enquanto o maior tingido corneal foi observado na zona inferior.

Também se observou que o uso de LC afetou as propriedades biomecânicas da córnea, no entanto, as diferentes propriedades não foram afetadas da mesma forma. O CRF diminuiu significativamente, verificando-se uma relação com o tipo de material das lentes; no entanto, não foram observadas alterações na CH. Verificou-se ainda que o ORA subestima a PIO medida sobre as LC e esta diferença ascendeu a 3 mmHg em cerca de um terço dos olhos avaliados; além disso, a PIOcc foi mais afetada do que a PIOg. Observou-se ainda que durante os três meses de uso de LC houve uma redução da PIO. Este efeito das lentes na PIO foi relacionado com o material da LC assim como com as propriedades corneais.

Este estudo permitiu observar que a SVC era maior com LC do que com óculos e que não sofreu nenhuma alteração durante os três meses de uso das lentes para nenhuma frequência espacial.

Em geral, este estudo mostrou que são várias as alterações que podem ocorrer na superfície ocular durante três meses de uso de LCH e que estas se encontram relacionadas com o tipo de material das lentes. A redução da DCC afeta a estabilidade do filme lacrimal o que pode levar a sintomas de secura ocular. As LC com elevada permeabilidade ao oxigénio também afetaram a fisiologia da superfície ocular induzindo hiperemia conjuntival e tingido corneal. No entanto, emboras estas alterações não sejam clinicamente significativas, podem afetar o nível de conforto dos utilizadores de LC. O efeito do uso de LCH na PIO deve ser tida em conta no tratamento de indivíduos com risco de glaucoma.

Uma vez que o mesmo material de lente afeta de forma diferente a citologia conjuntival, a fisiologia da superfície ocular ou a PIO, pode não haver uma LC ideal para todos os pacientes. Assim, o material das lentes deve ser selecionado para um determinado indivíduo dependendo da saúde ocular do mesmo.

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List of abbreviations

ATP adenosine triphosphate

CCLRU cornea and contact lens research unit

CH corneal hysteresis

CL contact lens

CLARE contact lens-induced acute red eye

CLPU contact lens-induced peripheral ulcer

CO_2 carbon dioxide

CRF corneal resistance factor

CRT corneal reshaping therapy

CS contrast sensitivity

CSF contrast sensitivity function

DAA diacetone acrylamide

Dk oxygen permeability

Dk/t oxygen transmissibility

DMA N, N-dimethylacrylamide

DNA deoxyribonucleic acid

EDTA ethylenediamine tetraacetic acid

EGDMA ethylene glycol dimethacrylic acid

EOP equivalent oxygen percentage

FDA food and drug administration

GAGs glycosaminoglycans

GCD goblet cell density

GM-CSF granulocyte-monocyte colony stimulating factor

GMA glyceryl methacrylate

GPC giant papillary conjunctivitis

H_2O hydrogen dioxide (water)

HCL hydrogel contact lens

HEMA hydroxyethyl methacrylate

HPMC hydroxypropyl methylcellulose

IFN interferons

Ig immunoglobulin

IL interleukin

IOP intraocular pressure

IOPcc corneal-compensated intraocular pressure

IOPg Goldmann-correlated intraocular pressure

LCP lens care product

LPCOFs lid-parallel conjunctival folds

MA methacrylic acid

MMA methyl methacrylate

MPa millipascal

MPDS multipurpose disinfecting solution

NIBUT non-invasive tear break-up time

NVA N-vinyl aminobutyric acid

NVP N-vinyl pyrrolidone

O_2 oxygen

ORA ocular response analyzer

PAS periodic acid schiff

PBVC poly(dimethysiloxy)iminocarboxyethyloxypropyl-poly(dimethylsiloxy)-butyldimethylsilane

PDMS polydimethylsiloxane

PEG polyethylene glycols

PHEMA polyhydroxyethyl methacrylate

PHMB polyhexamethylene biguanide

PMMA polymethyl methacrylate

PMN polymorphonuclear

PVA polyvinyl alcohol

PVP polyvinylpyrrolidone

RNA ribonucleic acid

SCL soft contact lens

SiHy silicone hydrogel

SLPI secretory leucocyte protease inhibitor

TBUT tear break-up time

TCA tricarboxylic acid

TPVC tris(trimethylsiloxysilyl)propylvinyl carbonate

TRIS tris(trimethylsiloxy)silane

UV ultraviolet

VMA N-vinyl-N-methylacetamide

CHAPTER 1.

MOTIVATION, OBJECTIVES AND STRUCTURE OF THE THESIS

1.1 Motivation

About 140 million people in the world are wearing contact lenses (CL) for the purpose of refractive error correction.[1] Among them, about 90% wear SCL.[2] Every year, millions of people commence to wear CL, however, the total number of CL wearers in the world has not been increased in the same ratio. This is because, other millions of already CL wearers discontinue to wear CL.[3] This indicates that, millions of CL wearers are not satisfied with the present CL, either with the lens materials or with the lens care system or with any unknown reasons. Studies on the effect of SCL wear on ocular surface are a well-established field. Many researchers are working in this field, some are devoting on the materials and correspondent properties, others in corneal surface, conjunctiva and others on eyelids or tears. However, there are still many areas for improvement. A good knowledge of the effect of CL wear on ocular surface helps clinicians to choose the most suitable CL for their patients and it will help the researchers to develop new materials with improved properties that induce minimum side effects. Currently various types of soft contact lenses (SCL) are available: from low permeable hydrogel lenses to hyperpermeable silicone lenses, water content ranging from 28-78%, various thickness and numerous other material characteristics. Every year several new CL with different materials and/or different parameters and properties come in the market. Because of these constant changes, clinical studies are necessary to determine the most suitable lenses for a particular type of patient.

The framework of this thesis is concentrating on the ocular surface which can be directly or indirectly involved on SCL wear. The research reported in this thesis is dedicated on the effect of CL wear either on corneal or on the conjunctival surface and on the eyelids. It was also studied the changes on tear function, which is the most anterior layer of the eye with a vital role on optical function and eye health. It was investigated the cytological changes on the conjunctival surface with CL wear. It was also examined the biomechanical properties of the cornea. Later on the thesis, it will be explained the effect of CL wear on contrast sensitivity (CS) and on intraocular pressure (IOP).

1.2 Objectives

The aim of the study presented in this doctoral thesis was to evaluate the changes in ocular surface induced by SCL wear. For the fulfilment of this purpose, it was conducted different clinical and experimental trials on more than 75 neophyte CL wearers. It was selected seven different types of recent CL with a wide range of materials, designs and parameters and two disinfecting solutions with different chemical composition.

One of the objectives was to determine the effect of SCL wear on conjunctival cytology. First, goblet cell density (GCD) in healthy eyes of non-CL wearers was determined and the relationship between GCD, tear function and ocular surface physiology was studied. Then, it was investigated the changes on conjunctival epithelial cell morphology and GCD with three months of SCL wear. Another objective was to investigate the changes in physiology of cornea and conjunctiva with lens wear. So, this experiment was designed to determine the effect of SCL wear on conjunctival bulbar and limbal redness and conjunctival and corneal staining. To determine the effect of SCL wear on corneal biomechanical properties, changes on corneal resistance factor (CRF) and corneal hysteresis (CH)] were also evaluated with the Ocular Response Analyzer (ORA). The objectives of the experiments on intraocular pressure (IOP) were to investigate the accuracy of IOP measurement over SCL and to determine the changes in IOP after three months of CL wear. For this experiment, it was measured the Goldmann-correlated intraocular pressure (IOPg) and corneal-compensated intraocular pressure (IOPcc) with ORA. The effect of CL wear on contrast sensitivity function (CSF) was evaluated with the CSV-1000 VectorVision. The actual trend of SCL is moving in the direction of daily wear modality. So, the other aim of the study was to compare the effect of SCL wear on ocular surface between daily and monthly wear modality.

1.3 Structure of the thesis

The present thesis is divided into eight chapters according to its main objectives: In the current **Chapter 1**, is presented, in short, the motivation and the main objectives of the study. A vigorous literature review regarding SCL, ocular surface and the effect of SCL wear on ocular surface is presented in **Chapter 2**. In brief, this chapter has been started with the history

of CL and the evolution of the different materials including the latest ones. Here, it was also explained the design, manufacturing process, their detail material composition specifically focusing on those lens materials which are used on the experiments of this study. Next section is focused on the anatomical and physiological aspects of the ocular surface. We mainly focused on those parts of eyes which are directly involved on CL wear. It was briefly described the anatomy and physiology of the cornea, conjunctiva, eyelids, limbus and tears. In the final part of this chapter, it is presented the effect of SCL wear on different parts of the eye. Since, both hydrogel contact lenses (HCL) and silicone hydrogel (SiHy) lenses were used during the experiments, any possible side effects of these lens materials are described with latest literature review.

As discussed in **Chapter 3**, it was performed a cytological clinical study on conjunctival tissues. In the first part of this experiment (3A), we have investigated the cytology on the bulbar conjunctiva of normal subjects who had never worn CL. In the second part of this experiment (3B), it was investigated the changes on conjunctival GCD and epithelial cell morphology after three months of SCL wear. **Chapter 4** presents the results obtained on the conjunctival and corneal physiology. It was determined the effect of three months of SCL wear on conjunctival redness, conjunctival and corneal staining.

As seen in **Chapter 5**, it was studied the effect of SCL wear on corneal biomechanical properties. Changes on CRF and CH with lens wear as well as the effect of lens materials on these changes were determined. **Chapter 6** consists of two parts related to SCL wear and IOP. In the first part, it was determined the accuracy of IOP measurement over SCL with ORA (6A). In the second part, it was investigated the effect of three months SCL wear on IOP (6B). The research on CS has been presented in **Chapter 7**. The difference on CSF between spectacles and SCL was determined initially and the CSF with SCL was also compared after three months of lens wear. The last chapter (**Chapter 8**) summarizes the whole thesis including major findings of this work, some limitations of the experiments performed in this thesis and finally highlighting some future works on the basis of the findings of this thesis.

References

1. Stapleton F, Keay L, Jalbert I, Cole N. The epidemiology of contact lens related infiltrates. *Optom. Vis. Sci.* 2007;84(4):257-72.
2. Morgan PB, Woods C, Tranoudis IG, *et al*. International contact lens prescribing in 2011. *Contact lens Spectr.* 2012;27(January):26-32.
3. Dumbleton K, Woods C, Jones LW, Fonn D. The impact of contemporary contact lenses on contact lens discontinuation. *Eye Contact Lens* 2013;39(1):93-9.

CHAPTER 2.

LITERATURE REVIEW

2.1 Contact lenses

2.1.1 History

Leonardo da Vinci sketched schematic eye and described the mechanism of image formation. He also described the ideas of vision improvement by using water-filled glass in contact with the cornea.[1] So, he is claimed as the first person to give the concept of CL which can be considered as the initial state of CL development. However, this is controversial.[2]

Sir John Herschel postulated the possibilities of correction of refractive error with a jelly or spherical glass over the cornea.[3] So, he can be considered as the father of CL. He also described the concept of prosthetic lenses.[4] Adolf E Fick is the first ophthalmologist to put CL of blown glasses on the cornea of a rabbit.[5] Later, these CL were filled with 2% glucose and put in irregular human corneas successfully. August Muller coined the term corneal lens. He corrected his own high myopia perfectly with CL. He also described the colored halos around lights due to corneal oedema.[4] In 1912, FA Muller and Albert Carl Muller who were glass-artificial-eye specialists, started to make CL of blown glasses which were very smooth and without a sharp edge. These lenses were more comfortable than Zeiss lenses which were produced by lathe-cut method. Lathe-cut lenses were better in vision.[6] In 1920, Carl Zeiss produced lathe-cut trial set which could be used for keratoconus eyes also.[6] Adolf A Muller-Welt started the first polymethyl methacrylate (PMMA) lens manufacturing in 1949.

2.1.2 Contact lens material developments

PMMA material was developed in 1934 by John Crawford and Rowland Hill at Imperial Chemical Industries.[7] Structure of PMMA is shown in figure 2.1. In 1936, PMMA was used in scleral CL by William Feinbloom.[8] Dennis C England prepared PMMA corneal lenses in 1946 but could not get patent.[9] But Kevin M Tuohy is the first scientist to get the patent on corneal PMMA lens in 1950. These lenses were 11 mm diameter and 0.4 mm thick.[9] These unicurve lenses should be fitted with 1.50D flatter. Later, George H Butterfield improved the lens with multicurve design.[10]

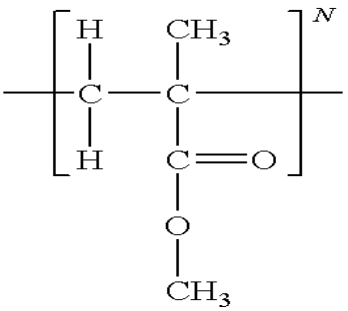

Figure 2.1 Structure of polymethyl methacrylate.

Otto Wichterle and Drashoslav Lim discovered polyhydroxyethyl methacrylate (PHEMA) lenses and trialed these spin-casting lenses in human cornea in 1956. PHEMA is a stable water absorbing polymer (with 38.6% water content) which is permeable to nutrients and metabolites (Figure 2.2).[9] Bausch & Lomb obtained FDA approval for Soflens in 1971.[11] Although, silicone elastomer lens was developed by Walter E Becker in 1956, it got FDA approval only in 1981 by Dow Corning. Bausch & Lomb bought this technology in 1985. John de Carle granted FDA approval to develop Permalens which is made up of HEMA, Vinyl pyrrolidone and methacrylic acid.[7] These lenses had 71% water and were used for extended wear.

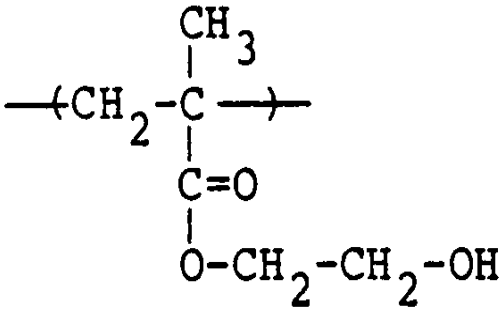

Figure 2.2. Structure of polyhydroxyethyl methacrylate.

Leonard Seidner is considered as the father of rigid gas permeable (RGP) CL. He, with Norman Gaylord developed RGP lenses made up of polysiloxanylalkyl acrylic ester and an alkyl acrylic ester and also a fluoroalkyl acrylic ester and a methacrylate material. Gaylord got a patent for Polycon in 1972. Then other higher oxygen permeable materials like fluorosilicones,

fluorocopolymers and polyethylene were developed.[9] Cellulose Acetate Butyrate lens was developed by Norman O Stahl and his group.[12]

Orlando A Battista developed the disposable lenses in 1978.[9] However, these lenses were made up of collagen and could be dissolved in some of the tear enzymes, so could not be successful commercially. Michael Bay developed commercially available disposable Danalens. However, they could not get success in the market.[9] The first successful disposable lenses were Acuvue® (Etafilcon A) from Vistakon®, Sequence® from Bausch & Lomb and NewVues® from CIBA Vision marketed in 1988. First daily disposable lenses were 1 Day Acuvue® by Vistakon®, Johnson & Johnson and Occasions® by Bausch & Lomb marketed in 1993.

SiHy lenses were introduced in the market in 1999.[13] Currently more than 15 SiHy lenses are available. These lenses contain silicone-oxygen bonds instead of carbon-carbon bonds in hydrogel lenses. Silicone-oxygen bond is longer in length, more mobile in comparison to carbon-carbon bonds, so SiHy lenses convey distinctive surface as well as mechanical properties.[14] They have high oxygen permeability, low frictional force, and low protein deposition but have a higher modulus and high lipid deposition. When introduced into the market, they have low water content and high contact angle ie low wettability.[14] During the last decade, the improvement in SiHy lenses occurred rapidly. Water content increased from 28% (Lotrafilcon-A) to 74% (Efrofilcon A), contact angle reduced (from > 40^0 in Lotrafilcon-A to <10^0 in Delefilcon-A), modulus reduced from 1.4 MPa to 0.3 MPa with little compromise in oxygen permeability.[14]

2.1.3 Material characteristics

Oxygen permeability

Oxygen permeability (Dk) is one of the most important characteristics of CL materials. The cornea is avascular, so oxygen required for the metabolic activities should be supplied through the atmosphere. SCL covers the entire cornea, so most of the required oxygen passes through the lens. So, lenses with sufficient Dk are necessary to maintain ocular health. Dk is expressed as:

$$Dk = Diffusion \times Solubility\ coefficient \qquad (2.1)$$

Diffusion coefficient (D) is the speed of oxygen molecules within the material (cm^2/sec) while k is the solubility coefficient of the oxygen in that particular material (mlO_2/ml mmHg).[15]

The unit of Dk is cm^2 mlO_2/sec ml mmHg and its value is in the order of 10^{-11}. The unit 10^{-11} cm^2 mlO_2 / sec ml mmHg can be denoted by barrer.[16]

Oxygen transmissibility

Oxygen transmissibility (Dk/t) is the permeability of oxygen per unit thickness (t). CL can be divided on the basis of Dk/t as shown in table 2.1. Dk/t can be measured in vitro by Colorimetric and Polarographic technique.[17,18] It can also be measured in-vivo by measuring equivalent oxygen percentage (EOP).

Table 2-1. Classification of soft contact lenses on the basis of oxygen transmissibility

Soft contact lens group	Oxygen transmissibility (barrer/cm)
Low	< 12
Medium	12-25
High	>25

Recently it has been postulated that oxygen transmission from the anterior surface of CL to the anterior surface of cornea depends upon the difference in oxygen tension in two sides.[19] The amount of oxygen moves from one side to the other side of CL is given by equation 2.2.

$$J = {Dk(P_0 - P_1)}/{t} \qquad (2.2)$$

Where *J* is the oxygen flux, Dk is oxygen permeability and t is the thickness of the CL, P_0 is the oxygen tension on the anterior surface of CL and P_1 is the oxygen tension on the posterior surface of CL. At lower transmissibility, the oxygen flux increases rapidly with increase in Dk/t, but after it reaches the certain point, it increases very slowly on increasing Dk/t.

Water content

It is an important characteristic of lens materials. Lenses with high water content are very hydrophilic and difficult to handle while lenses with low water content are less oxygen permeable (in cases of hydrogel lenses) and low ion permeable. It also determines the comfort level of the lenses. The water content of the SCL is expressed as a percentage of a total lens once it is hydrated

fully in normal saline. In non-silicone hydrogel lenses, oxygen passes through the lens in dissolved form, not by gaseous form. Some of the water in hydrogel lens material exists as charged molecule or dipole which is electrostatically bound with the polymer by hydrogen bond. This bound or non-freezing water has not a role in oxygen permeability.[20] The other portion of water which is without charge, also called freezing water, involves in oxygen transport. So, the amount of freezing water is important for Dk/t rather than total water content in HCL. High water content lens transmits higher oxygen but if the thickness of high water lens is low, pervaporation of the cornea takes place.[21] Oxygen passes through the lenses forming a bond with silicone molecules in SiHy lenses. So, the higher the silicone component, the higher will be the Dk.[14] In most of the silicone lenses, the lower the water content, the higher the oxygen transmission.[22] Figure 2.3 shows the relation between water content and Dk in HCL and SiHy lenses. This relation is different with other latest non-Tris SiHy lenses.[23] In SiHy lenses, the water content is also important for ion transport. A minimum of 0.2 X 10^{-6} cm^2/s of ion transport is necessary for normal ocular health.[24]

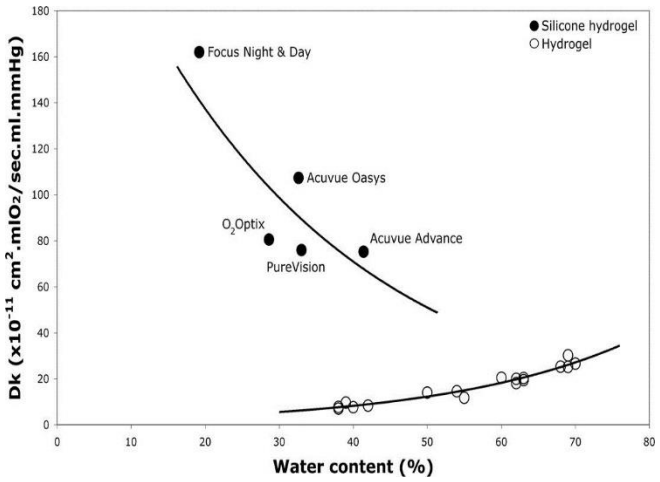

Figure 2.3. Relation of oxygen permeability and water content in hydrogel lenses and silicone hydrogel lenses. Reprinted from Efron N, Morgan PB, Cameron ID, Brennan NA, Goodwin M. Oxygen permeability and water content of silicone hydrogel contact lens materials. *Optom Vis Sci* 2007;84(4):328-337[25] with permission from the Wolter.

Food and Drug Administration (FDA) of USA classify SCL according to the water content (high with more than 50% and low with less than 50%) and the presence of ionicity on the lens surface (ionic or non-ionic) (Table 2.2).[26]

Table 2-2. FDA classification of conventional soft contact lenses.[26]

Group	Water content	Net surface charge
I	Low	Non-ionic
II	High	Non-ionic
III	Low	Ionic
IV	High	Ionic

However, with SiHy lenses, given their unique chemistry, the classification does not necessarily perform as predicted. SiHy lenses exhibit different pattern of deposition and wettability properties. Hutter et al. proposed a new system of FDA classification as shown in table 2.3 where lens materials are classified based on water content, surface ionicity, surface treatment and monomer characteristics.[27]

Table 2-3. Purposed FDA classification of silicone hydrogel lenses [adapted from Hutter *et al.*][27]

Group	Water content	Net surface charge	Surface treatment	Monomer characteristic
VA	Low	Non-ionic	Yes	Any
VB1	Low	Non-ionic	No	Hydrophilic
VB2	Low	Non-ionic	No	Semi-interpenetrating network
VC	High	Non-ionic	Yes or no	Any
VD	High and low	Ionic	Yes or no	Any

Wettability

CL wettability is the ability of the tear film to cover and maintain itself over CL surface[28] and is considered as an important factor in determining its physiologic compatibility.[29] It also affects lens deposition,[24] optical quality and comfort.[30] The wettability of a CL is dependent on the surface tension of the tears, the free energy of the CL and the interfacial tension between them.[31] Lipid layer of the tears lowers surface tension and slows evaporation.[32] When a CL is placed on the eye, the surface chemistry can be changed due to tear composition and different environment.[33] Lower the surface tension of the tear film, better the wettability of the lens.[31] CL

may disrupt the tear layer and the precorneal tear film of about 4 µm becomes prelens tear film of about 2.5 µm.[34-36] Due to CL wear, it interrupts tear film reformation,[34] increases tear osmolarity[37,38] and degrades the lipid/mucin which reduces the CL wettability.[39] Although the main purpose of the surfactants present in modern lens care system is to remove deposits, they help in improving wettability of CL by reducing the surface tension of tear film and increasing viscosity.[40]

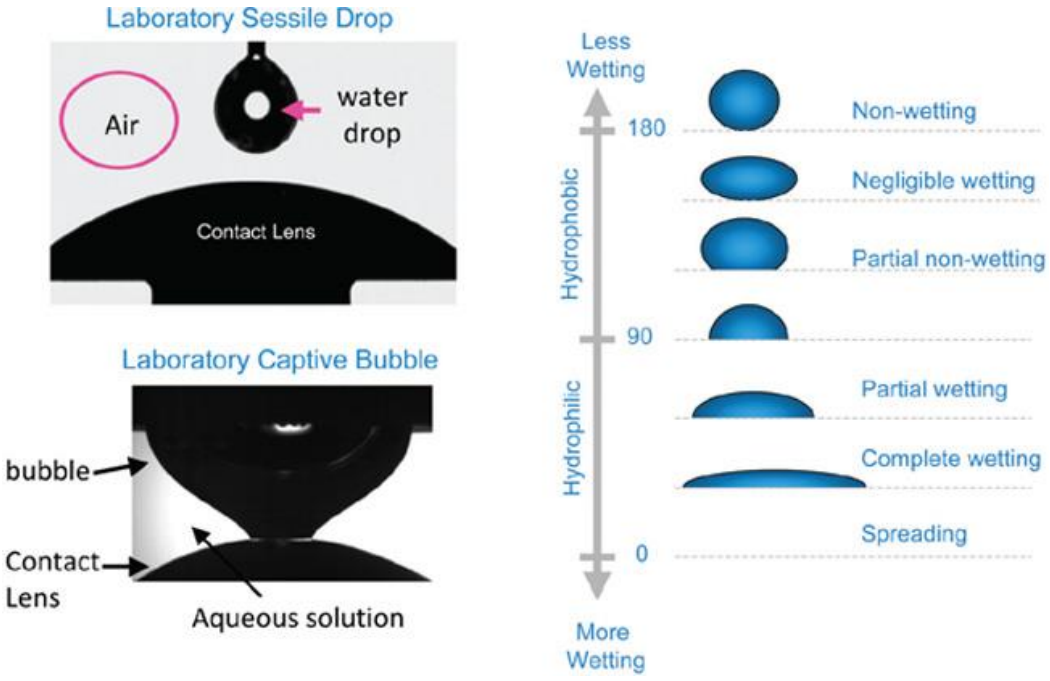

Figure 2.4. Techniques of contact angle measurement and the relationship of the contact angle with wettability. The sessile drop method measures the angle between a drop of water and a contact lens surface exposed to air. Dynamic captive bubble technique measures the angle between an air bubble and a contact lens surface in an aqueous environment. Reprinted with permission from the 2011 special edition of Contact Lens Spectrum, published by Pentavision LLC. © 2011 All right reserved.[41]

Different technologies have been used to enhance the wettability of current CL. Water gradient surface has been applied on Delefilcon A lens in which water content of the core is 33% while on the surface is more than 80%.[30] In AquaGen technology, hydrophilic monomers migrate to the lens surface to increase lens wettability such as in Comfilcon A and Enfilcon A lenses.[30] Internal wetting agents with long chain, high-molecular-weight chemicals based on polyvinylpyrrolidone are used in some SiHy lenses (MeniSilk technology) like Narafilcon A,

Galyfilcon A and Senofilcon A.[30] Similarly, some blister pack solutions contain surfactants like polyethylene glycon, hydroxypropyl methylcellulose, polyvinyl alcohol, polyvinylpyrrolidone which also enhance the wettability of the lenses.[30] Surface wettability can be estimated by measuring contact angle [wettability and the contact angle are not the exact same thing[42]]. Contact angle is the angle between contact lens and liquid interface and it is the indication of the level of attraction between the material and the liquid.[43] The smaller the contact angle, the greater the attraction which means better wettability (Figure 2.4). Lens surface wettability can be measured in vivo (*e.g.*: tear coverage, break-up time, drying time measured on slit-lamp) or in vitro (Sessile drop, Wilhelmy plate or Captive bubble) methods. Generally, a contact angle of less than 90^{O} is wettable. Receding angle is always less than advancing angle. However, in captive bubble method, the receding angle is larger than advancing angle.[30]

Modulus

Modulus shows the flexibility or the stiffness of a material. It is a measure of how a lens can deform under pressure. It is an important physical property of the CL material as it determines the mechanical impact on the ocular surface. Lens with low modulus is difficult to handle and less durable while a lens with high modulus can induce mechanically induced ocular pathology. Higher modulus lenses are more durable, easier to fit and mask some anterior corneal astigmatism. Generally, silicone lenses have high modulus in comparison to hydrogel lenses because of the stiffness of the silicone material. The flexibility of lens can be measured in vitro by measuring residual astigmatism.

Optical quality

Optical quality of the CL materials should be good. It must be transparent and homogenous in refractive spectral transmission. Inhomogeneity in optical quality may result in scattering of light. A good quality material has little or absence of dispersion and chromatic aberration. The refractive index is measured with green mercury light (546.07 nm) while dispersion is the amount of difference in refractive index with blue (479.99 nm) and with red (643.85 nm) light.[44]

Biocompatibility

Biocompatibility is the most important property of CL as correspond to the ability of the material to perform its intended function with the desired degree without any undesirable side

effects.[23] Because CL is placed directly on the cornea, it should be inert with ocular tissue as well as lens care products. It should not absorb any metabolites, toxins, micro-organisms and should be electrically compatible. It should not induce an inflammatory or immunological response to ocular surface. Modern CL are not fully biocompatible to the ocular tissues so that there is still some chance of complications due to CL wear.[45,46]

2.1.4 Soft contact lens materials and manufacturing process

Polyhydroxyethylmethacrylate (PHEMA) is the first material used to make SCL. It has a polar hydroxy group and contains 38% water. In 1968 Griffin developed another SCL material (Bionite Naturalens) which is the mixture of PHEMA and Polyvinylpyrrolidone (PVP) that contains 55% water e.g. Vifilcon A. Most of the current CL are made up off co-polymer or ter-polymer (two or three different polymer linked by a cross-linking agent) of following monomers: PHEMA, PVP, methacrylic acid (MA), methyl methacrylate (MMA), glyceryl methacrylate (GMA), diacetone acrylamide (DAA) and polyvinyl alcohol (PVA). Generally used crosslinker is ethylene glycol dimethacrylic acid (EGDMA) which helps the polymer to increase dimensional stability.[47] Inclusion of MA makes lens negatively ionic. Ionic materials are more wettable, and denature protein less but they deposit higher charged particle like positively charged lysosomes and susceptible to pH change.

SCL are manufactured by molding, spin-casting or lathing. In molding, monomers are mixed and then poured into a mold in controlled environment. Ultraviolet (UV) radiation is necessary to initiate polymerization. Later, it is hydrated, polished and refined. Light stream manufacturing process includes reusable molds and makes fully hydrated lenses so that further extraction steps are not necessary. A controlled stream of UV light helps in cross-link.[48] Photolithographic process is used to make an edge (an opaque mask is kept on the periphery to prevent the cross-linking on the edge). Because of reusable molds which are made up of high-quality quartz and glass, the production cost is cheaper. Spin-casting is a method where an open-backed mold is spun as a small centrifuge. The mold defines the front surface and the rotational velocity, surface tension and gravity define the back surface. These lenses are softer. In lathing-xerogel, an anhydrous button of the lens material is lathed in a controlled atmosphere. It is then hydrated, autoclaved in 121 degrees for 15 minutes and packed in solution for marketing. These lenses are less flexible. SCL can also be manufactured by a combination of molding, lathing and

spin-casting. Recently, a stabilized-molding technique has been introduced. In this new technique, a space-taking inert diluent is included in the mixture of monomers during molding/polymerization. Later, the diluent is replaced by water. This provides high-quality optics with high reproducibility lenses. Lenses are packaged in glass vial or polyethylene terephthalate vial with a screw or crimp lid. Disposable lenses are packaged in a foil pack or multi-blister pack. Before marketing, these are autoclaved.

Materials and other characteristics of some current lenses

Silicone elastomer lenses were manufactured during the 1980s, for example, Silsoft of Bausch & Lomb. The homopolymers of siloxane such as polydimethylsiloxane (PDMS) have high Dk. Because of their hyper Dk, they were indicated for 30 days extended wear and become more popular especially in aphakic children.[49] However, because of their hydrophobic nature, low water content, poor lens wetting and rapid lipid deposition, they can not be used furthermore and discontinued from manufacturing.[50] In 1999, SiHy lenses came into the market with the introduction of Balafilcon A by Bausch & Lomb and Lotrafilcon A by CIBA Vision.[51] Material characteristics of the studied lenses are presented in table 2.4.

Lenses with material Balafilcon A (Purevision2^{TM}) contain tris(trimethylsiloxy) silane (TRIS) and N-vinyl pyrrolidone (NVP). As shown in figure 2.5, TRIS, the silicone-containing monomer helps in oxygen permeability while NVP helps in fluid and ion transport. It took such a long time to develop SiHy lenses because it is tough to mix TRIS with NVP, these oil-and-water like materials can lose transparency on mixing. Since the surface was very hydrophobic, plasma oxidation was done. Plasma oxidation in a gas chamber converts TRIS structure on the surface into hydrophilic silicate. Balafilcon A has 1.1 MPa modulus and 36% of water content (Table 2.4).

Lotrafilcon B (AirOptix® Aqua^{TM}) is made by polymerizing TRIS macromers with silicone elastomer sequences interspersed with hydrophilic polyethylene glycols (PEGs). It has biphasic or two-channel molecular structure that is formed by a fluoroether macromer copolymerized with a TRIS monomer and the hydrophilic monomer N, N-dimethylacrylamide (DMA). Fluorosiloxane phase allows high oxygen permeability while the hydrogel phase helps in ion and water movement. It has fine phase separation (size of phase separation is less than the light wavelength) to maintain the optical clarity. Plasma treatment is done which creates a permanent

ultrathin (25 nm) hydrophilic layer on the surface hiding silicone structure. It has a modulus of 1.2 MPa less than Lotrafilcon A (1.4 MPa). Lotrafilcon B has higher water content and lower Dk than Lotrafilcon A but other chemical properties are similar.

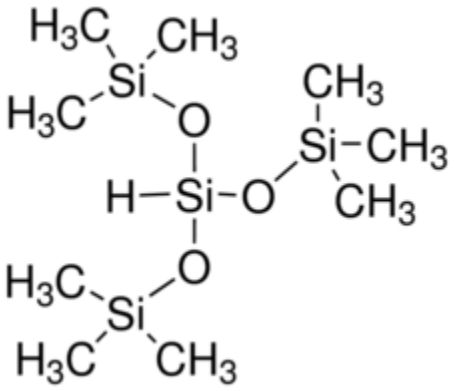

Figure 2.5 Tris(trimethylsiloxy)silane (TRIS) structure

Comfilcon A (Biofinity^{TM}) uses a unique long-chain siloxane macromer combined with other monomers to obtain relatively low modulus and high oxygen permeability. It does not have any surface treatment and internal wetting agent. Silicone component is the source of oxygen permeability and Comfilcon A has 48% water and 128 barrer Dk. It has a modulus of 0.75 MPa (Table 2.1).

Nelfilcon A (Dailies® AquaComfort Plus®) is composed of modified polyvinyl alcohol 31% and water 69 %. It has 26 barrer Dk and is in group II (non-ionic, high water) of FDA classification.[48] The lens is tricurve design with front optic zone diameter of about 8.00 mm and edge thickness is 75 µm. The lenses are cross-linked in the molds by controlled exposure to ultraviolet light. This light stream process eliminates the need of further time and money consuming cleaning process.

Table 2-4. Material characteristics of the study lenses.

	Dailies® AquaComfort Plus®	Biotrue™ ONEday	1-Day Acuvue® True-Eye™	AirOptix® Aqua™	Biofinity™	PureVision2™	MyDay™
Company	Alcon Vision Care	Bausch & Lomb	Vistakon (Johnson & Johnson)	Alcon Vision Care	Cooper Vision	Bausch & Lomb	Cooper Vision
Material (USAN name)	Nelfilcon A	Nesofilcon A	Narafilcon A	Lotrafilcon B	Comfilcon A	Balafilcon A	Stenofilcon A
Water content (%)	69	78	46	34	48	36	54
Wearing modality	Daily disposable	Daily disposable	Daily disposable	Monthly disposable, daily wear	Monthly disposable, daily wear	Monthly disposable, daily wear	Daily disposable
FDA Classification	II	II	I	I	I	III	
Thickness (mm) – 3.00D	0.10	0.1	0.085	0.08	0.08	0.07	0.07
Permeability (Barrer) (-3.00D)	26	42	100	110	128	99	80
Modulus (MPa)	0.89	0.49	0.66	1.2	0.75	1.1-1.25	0.4
Principal monomers	Not disclosed	HEMA & NVP	PVP	DMA+TRIS +Siloxane monomer	FM0411M, HOB, IBM, M3U, NVP, TAIC, VMA	NVP+TPVC +NVA+PBVC	Not disclosed

[DMA = N, N-dimethylacrylamide, FM0411M = α-meth**acryloyxyethyl iminocarboxyethyloxypropyl-poly(dimethylsiloxy)-butyldimethylsilane,** HEMA = hydroxy ethylmethacrylate, HOB = 2-hydroxybutyl meth**acrylate, IBM = isobomyl methacrylate,** TRIS = trimethylsiloxy silane, M3U = α ώ-Bis(meth**acryloyloxyethyl iminocarboxy ethyloxypropyl)-poly(dimethylsiloxane)-poly(trifluoroprypylmethylsiloxane)-poly(ώ-methoxypoly(ethyleneglycon)propyl methylsiloane), NVA = N-vinyl aminobutyric acid,** NVP = N-vinyl pyrrolidone, **PBVC = poly(dimethysiloxy)iminocarboxyethyloxypropyl-poly(dimethylsiloxy)-butyldimethylsilane, TAIC = 1,3,5-triallyl-1,3,5–triazine-2,4,5(1H, 3H,5H)-trione,** TPVC = tris-(trimethylsiloxysilyl) propylvinyl c**arbonate, VMA = N-Vinyl-N-methylacetamide**].

Stenofilcon A (MyDay™) is one of the latest daily disposable silicone hydrogel lenses launched in June 2013 in the European market. It is made by Smart Silicone chemistry. This technique makes the silicone into a network of channels which help in oxygen delivering. It does not contain any surface treatment or internal wetting agents.

Nesofilcon A (Biotrue™ ONEday) is a daily disposable hydrogel lens which is manufactured with a new material called a "HyperGel" which is a copolymer of HEMA and N-vinyl pyrrolidone.[52] It has 0.49 MPa modulus and UV blocking potential and packaged in a borate buffer solution with poloxamine. This lens is very good resistant to dehydration.[52]

Narafilcon A (1-Day Acuvue® TruEye) is the first daily disposable silicone lenses available in the market. It contains polyvinyl pyrrolidone (PVP) as internal wetting agent via Hydralcare-1 technology. It is class 1 UV blocking lens which blocks more than 96 % of UVA and 100 % of UVB.

2.1.5 Lens care system

SCL should be kept on the aqueous state to maintain its hydration. New CL are kept in blisters with saline solution by the manufacturers to maintain hydration and tonicity. All the soft lenses except daily disposable lenses require a solution to clean and soak them after wear and to store and keep them disinfected while they are not being used. CL solution helps the lenses to maintain hydration and keep lenses out of micro-organisms.[53] The ideal CL solution should be compatible to the ocular surface with efficient disinfectant properties and should be able to maintain hydration of the lenses.[53] After the mid-1980s, multipurpose disinfecting solutions (MPDS) were introduced in the market with the aim to do all the functions by a single solution.[23] Nowadays, more than 90% of soft CL wearers use MPDS.[54,55] A typical MPDS contains disinfectants, surfactants, buffers, salts, chelating agents and wetting agents. Recently, after the global outbreak of Fusarium keratitis (with the use of ReNu MoistureLoc, Bausch & Lomb) and *Acanthamoeba* keratitis (with the use of Complete MoisturePlus, Advanced Medical Optics) in 2005 to 2007, MPDS with double disinfectants have been used.[56]

Disinfectants should be able to kill micro-organisms especially the common micro-organisms that affect ocular surface like *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Serratia* marcescens, *Candida albicans* and *Fusarium solani*.[53] The international organization for

standardization standard 14729 standalone test limit on the ability of MPDS to kill at least 3 log unit of bacteria and 1 log unit of fungi, but it does not say anything for *Acanthamoeba*.[57] Because these chemicals can affect the ocular surface, they are used in low concentration. They should not bind or absorb on the lens surface since these components can be released during wear and induce reactions on the ocular surface. Chlorhexidine and thimerosal are good disinfectants because of their strong disinfectant property, however, hydrogel lenses adsorb (attachment onto the lens surface) or absorb (move into the lens matrix) these low-molecular weight preservatives.[58] They ultimately release on the eye during lens wear and induce allergic reaction. So these disinfectants are not used in current CL solutions.[45] Most of the current disinfectants found in CL solutions have higher molecular weight, which cannot penetrate the lenses becoming less reactive. Common disinfectants found in current CL solutions are Polyhexamethylene biguanide HCl (PHMB), Polyquaternium-1, Myristamidopropyl dimethylamine (Aldox), polyaminopropyl biguanide, Alexidine and hydrogen peroxide.

PHMB is a larger, highly cationic, relatively hydrophilic biocide which is a good disinfectant that works by binding with negatively charged plasma membrane of the microbes that causes membrane disruption and cellular damage.[53,59] Polyquaternium-1 (Polyquad) also acts on pathogen plasma membrane.[60] Because of its high molecular weight, it cannot penetrate hydrogel lenses and works with lower concentration.[60] Aldox (Myristamidopropyl dimethylamine) is a small, cationic agent which is relatively hydrophobic biocide acts on both antifungal and antiamoebic organisms.[53,60] Alexidine is a cationic bibiguanide[53] which is similar with chlorhexidine and is considered that it acts with lower concentration.

Surfactants/cleaners introduced in MPDS help to remove lens deposits. They are organic compounds with amphipathic nature having one hydrophilic and other hydrophobic end. Hydrophobic end interacts with hydrophobic lens surface while hydrophilic region interacts with hydrophilic tear film reducing surface tension and enhancing lens wettability.[30] They remove loose debris and deposits and enhance wettability of the lenses.[53] Poloxamers (Pluronic), poloxamines (Tetronic), isopropyl alcohol, sodium citrate, sodium borate, sodium phosphate, polyvinyl alcohol are some currently available surfactants.[53,61] CL solutions also contain some chelating agents which enhance the efficiency of disinfectants and also help in removing tear proteins.

Ethylenediamine tetraacetic acid (EDTA), citrate and hydroxyalkylphosphonate (Hydranate) are common chelating agents.[53]

Lubricants are also present in some MPDS. They act by attracting moisture. Commonly found lubricants include hydroxypropyl methylcellulose, hydroxypropylguar, tetronic, dexpanthenol, sorbitol and poloxamine.[61] Moreover, some water soluble polymers like hydroxypropylmethylcellulose (HPMC) or propylene glycol are also found in some current CL solutions as demulcent which relieve dryness and irritation and improve comfort.[53] To maintain the tonicity of a solution, salts are added in the solution. A common salt used is sodium chloride.[61] Similarly, buffers like borate, phosphate, nitrate, bicarbonate and citrate are used to control the pH.[53]

Hydrogen peroxide is considered as "gold standard" disinfectant in CL solution.[53] A 3% concentration (30,000 ppm) is used. However, to avoid its cytotoxic effect to ocular tissues, it is neutralized to 100 ppm before in contact with ocular surface.[53] Because, it can penetrate the lenses, it cleans lenses thoroughly by expanding the lens matrix and oxidizing microbes. Due to its low pH of 4.00, it can break protein and lipid bonds and remove trapped debris and so eradicates microbial biofilms.[61] Research show that use of another MPDS induces ten times more corneal infiltrates than the use of hydrogen peroxide for lens disinfection.[56]

Table 2.5 shows the components of the solutions used in the present study.

Table 2-5. Ingredients of contact lens solutions used in the current thesis.

	OPTIFREE® Puremoist	AOSEPT® Plus
Disinfectants	Polyquaternium-1 (0.001%), Aldox (0.0006%)	Hydrogen peroxide (3%)
Wetting agents	Poly(oxyethylene)-poly(oxybutylene) ie HydraGlyde	
Chelating agent	EDTA	
Others	Tectronic 1304, Sodium citrate, Sodium chloride, Boric acid, Sorbitol, Aminomethylpropanol	Phosphonic acid, sod. Chloride, phosphate, poloxamer

2.2 Ocular surface anatomy and physiology

2.2.1 Cornea

The cornea is one of the most important parts of the eye in relation to vision. It forms a smooth refractive component of the eye together with tears. The shape of the cornea is meniscus concave with posterior curvature steeper than that of anterior curvature so that its central thickness is less than that of peripheral region.[62] Cornea flattens towards periphery in an asymmetric way creating an aspheric optical system.[63] The average corneal thickness in an adult eye is approximately 539 to 544 µm in the center and 626 to 652 µm in the periphery.[64,65] It is highly hydrophilic in nature containing more than three-fourths water. Cornea transmits about 97% of the light in the visible spectrum.[66] It provides protection from the invasion of micro-organisms. This is possible due to the balance in corneal anatomy and physiology.[66,63]

Cornea is made up of five layers as shown in figure 2.6.[67] Epithelium, stroma and endothelium are cellular layers while Bowman's layer and Descemet's membrane are interfaces.[63] All the layers can be seen using a slit lamp biomicroscope with different magnification and illumination intensity and observation methods.

Epithelium is the outer surface of the cornea supporting the tear layer.[62] It is one of the most important layers because it acts as main refractive interface together with tear film. Epithelium with tear interface provides two-thirds of the total refractive power of the eye.[63] It also forms the barrier to the outside environment. The thickness of the epithelium is 48-60 µm and is comprised of five to seven layers which are made up of three types of cells.[68] Two to three layers of squamous cells are found in the surface which are regular in size and thickness. The middle layer is composed with 2-3 layered wing cells. The innermost single cell columnar layer is formed by basal cells which are attached to the underlying basement membrane by the hemidesmosomal system. Basal cells are regular in non-CL wearers or highly gas permeable lens wearers but found irregular in SCL wearers with extended modality.[69] Peripheral basal cells contain epithelial stem cells which are capable for mitosis and are a source of wing cells and surface cells. From one to two weeks, these cells complete the journey of migration to wing cells then to squamous cells and then exfoliation from the ocular surface.[70] Macrophages and lymphocytes are present on the basal

layer. Pigmented melanocytes are also present in the peripheral region of the basal cells. Regular replacement of the epithelium (apoptosis and desquamation) takes place with cellular turnover from basal to surface which takes 7-10 days for a complete turnover.[63,62] Photo Electron Micrograph of epithelium shows microplicae and microvilli on the surface layer covered with glycocalyx layer which plays important role in tear film stability by anchoring the mucus layer.[62] Tear film protects the epithelium from microbial invasion, from chemicals or toxins and foreign body damage and also provide immunological and growth factors which are necessary for epithelial cell health, proliferation and repair.[63]

Between epithelium and bowman's layer, there is basement membrane also known as basal lamina with thickness 10-65 nm.[62] It comprises type IV collagen and laminin secreted by epithelial cells.[63] Its anterior surface is well defined while posterior surface blends with Bowman's layer.

Bowman's layer is an acellular tissue with thickness 8 to 16 µm and contains dispersedly arranged collagen fibrils and some ground substances.[68,65] Collagens are smaller and less densely packed in comparison to that of the stroma. If disrupted, Bowman's layer cannot regenerate but develops a scar.[63]

Stroma is the main part of the cornea comprising approximately 90% of the total corneal thickness.[71] Central thickness of the stroma is about 478 µm and peripheral thickness is about 584 µm.[65] It has an important role in transparency and mechanical strength of the cornea. It is composed by collagenous lamellae and a small amount of keratocytes (corneal fibroblast) (2-3%) and glycosaminoglycans (GAGs) (1%). Stroma contains type I and V collagen.[72] Collagen fibers are arranged in parallel bundle and form fibrils. Many fibrils are packed in regular fashion forming lamellae. Stromal lamellae are fibrous connective tissue which are densely and orderly arranged. They are parallel with each other and also with cornea.[62] Transparency of the cornea is due to the fact that light scatter is eliminated by destructive interference of the stable protenous collagenous fibrils. GAG is very hydrophilic and responsible for the H_2O inhibition pressure of the cornea. Keratocytes are found in between collagenous lamellae which are thin and flat cells with about 10 µm diameter with long cytoplasmic processes and with 5-55 µm intercellular space.[73] In normal eyes, the density of keratocytes is about 20,000 cells per square millimeter.[65] These rough-surfaced endoplasmic reticulum like cells are joined with each other by macula occludens or hemidesmosomes. These cells synthesize collagen molecules and GAGs.

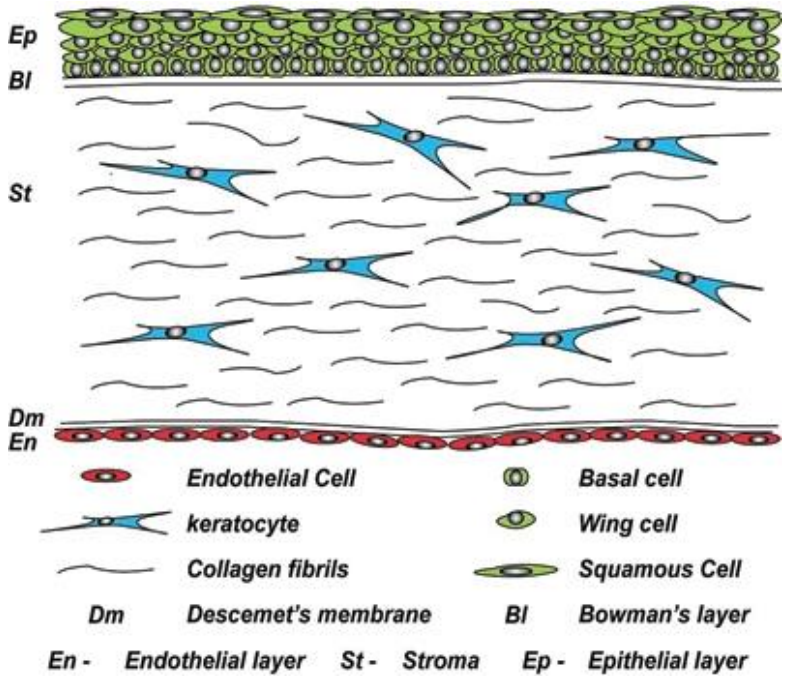

Figure 2.6. Transverse section of the cornea. [adapted from https://commons.wikimedia.org/wiki/File:The_human_cornea_in_cross-section..jpg, assessed on 30 August 2015]..

Stroma is the main part of the cornea comprising approximately 90% of the total corneal thickness.[71] Central thickness of the stroma is about 478 µm and peripheral thickness is about 584 µm.[65] It has an important role in transparency and mechanical strength of the cornea. It is composed by collagenous lamellae and a small amount of keratocytes (corneal fibroblast) (2-3%) and glycosaminoglycans (GAGs) (1%). Stroma contains type I and V collagen.[72] Collagen fibers are arranged in parallel bundle and form fibrils. Many fibrils are packed in regular fashion forming lamellae. Stromal lamellae are fibrous connective tissue which are densely and orderly arranged. They are parallel with each other and also with cornea.[62] Transparency of the cornea is due to the fact that light scatter is eliminated by destructive interference of the stable protenous collagenous fibrils. GAG is very hydrophilic and responsible for the H_2O inhibition pressure of the cornea. Keratocytes are found in between collagenous lamellae which are thin and flat cells with about 10 µm diameter with long cytoplasmic processes and with 5-55 µm intercellular space.[73] In normal eyes, the density of keratocytes is about 20,000 cells per square millimeter.[65] These rough-surfaced endoplasmic reticulum like cells are joined with each other by macula occludens or hemidesmosomes. These cells synthesize collagen molecules and GAGs.

Descemet's membrane is a structureless elastic membrane with thickness about 10-12 µm which is secreted by endothelium.[63] it is produced by the endothelial cells and can be regenerated if injured.[71] It is the most resistant part of the cornea to trauma and disease.[71] Periodic thickenings of the Descemet's membrane (Hassall-Henle Warts) can be found protruding anterior chamber covered by thin endothelium.

Endothelium is the innermost layer of cornea containing a single layer with about 500,000 hexagonal cells with diameter 18 to 20 µm and 5 µm thickness.[62] These cells cannot replicate once they are destroyed.[71] In a healthy eye, nuclei of the endothelium are flattened and centrally located. Due to its non-replicating nature, loss of cells by different circumstances or by age-related degeneration results decrease in uniformity of thickness, decrease in cell density and increase in the variation of cell sizes (polymegethism) as well as an increase in cell size (pleomorphism). Polymegethism is common in long-term extended wear low Dk lens wearers.[74] The density of endothelial cells is 3500 cells /mm^2 at birth which is decreased by 0.6 cells/mm^2 every year.[63] Patel *et al.* did not find a difference in endothelial cell density in CL wearers and non-CL wearers.[65] Endothelium contains a large number of mitochondria, mostly around the nucleus. It involves in active transport for protein synthesis and hydration control.

Cornea is avascular in nature, however, the peripheral part receives blood supply by conjunctival, episcleral and scleral branches of circumcorneal vessels which play a minor role in corneal nutrition. Cornea is supplied by the ophthalmic branch of fifth cranial nerve (trigeminal nerve). Because of the absent of myelin sheath in corneal nerves, nerve fibers are not visible. But nerve fibers are more visible in the oedematous cornea. Deep layers like Descemet's membrane have very few nerve supply. Cornea gets sensory, parasympathetic and sympathetic innervation.

Corneal transparency

Cornea is optically transparent comprising two-thirds of the total refractive power of the eye. Collagen fibrils are regularly arranged on the stroma which is the main part of the cornea. Light transparency is possible because the irregularity of collagen fibrils does not exceed a few wavelengths. Swelling disturbance the arrangement of fibrils and causes scattering effect and loss of transparency.[62]

Corneal swelling occurs due to lactic acid accumulation or due to high water retention by hydrophilic GAGs. During sleep, cornea swells because of hypoxia followed by anaerobic glycolysis (50%) and due to decreasing in osmolarity of tears so that water moves into the cornea and also increases in temperature and humidity (50%). Corneal swelling, especially an oedematous basal layer of epithelium, alters the refractive index of intracellular materials which causes Sattler's veil and haloes. However, even with the corneal swelling, corneal curvature is maintained.[75]

Endothelial pump which is an active Na^+ K^+ + ATPase-dependent glucose-fueled pump, plays a minor role in recovery from corneal oedema (about 20%). Each cell pumps its own volume every five minutes. Epithelial pump is an active pump where chloride ion from tears enters into cornea and sodium ion from cornea moves into tears.[76] Lactate generated here is moved posteriorly towards aqueous humor. Stromal pump is relatively inactive except keratocytes metabolism. Thus cornea thickens due to hypoxia or reflex tearing (hypotonic tears) while thins due to osmotic response (hypertonic). About 3-4% corneal swelling on closed-eye condition during sleep is normal which can normalize within few hours of eye opening.

Metabolism

Corneal endothelium has greater water permeability than the epithelium. The majority of the oxygen required for cornea is obtained from the atmosphere while little by palpebral conjunctiva and limbal vasculature (in closed eye circumstances). The cornea is highly permeable to CO_2 (7 times than that of O_2). Sodium ion is highly permeable to endothelium (100 times than epithelium) and glucose and amino acids are also permeable.

Epithelium is low permeable to Na+. It is relatively impermeable to water, glucose, amino acids, lactic acid and large molecules but permeable to fat-soluble entities. Cell junctions help in intercellular communication and act as a barrier to electrolytes, fluids and macromolecules. Mainly two types of junctions are found in corneal epithelial cells: tight or occluding junction (zonulae/belts or fasciae/band or maculae/focal). A hemidesmosome is a special type of desmosome found in the basal cell layer of the corneal epithelium which anchor the epithelium to its basement membrane and sometime may reach or even penetrate Bowman's layer. Cornea synthesizes fibronectin molecules which help in cell adhesion.

Gas movement to the eyes

Epithelial surface obtains oxygen from the atmosphere (20.9% or 155 mmHg partial pressure) during open eye condition and from conjunctival and limbal arteries (7.7% oxygen tension on the central cornea) during closed eye condition. To avoid the corneal oedema, a partial pressure of 10.1% of the oxygen is necessary.[77] Stroma and endothelium derive oxygen from aqueous humour (7.4% or 55 mmHg). CO_2 from the cornea is passed out (efflux) to the atmosphere during open eye and through aqueous during closed eye condition. In normal open eye condition, oxygen flux is 2-11 $\mu L.cm^{-2}.h^{-1}$.[78,79] Oxygen uptake rate with SCL during open eye condition is also similar with these values ranging from 2-10 $\mu L.cm^{-2}.h^{-1}$.[80] However, oxygen flux into the cornea does not indicate the exact rate of oxygen consumption.[19] Corneal oxygen consumption rate increases linearly until the Dk/t of 15 barrers/cm during open eye condition and until 300 barrers/cm during closed eye condition; after that, oxygen consumption rate flattens with increase in Dk/t.[81] The maximum oxygen consumption rate of cornea is found 4.5 X 10^{-5} mL.cm $^{-3}.s^{-1}$ to 22 X 10^{-5} mL cm^{-3} s^{-1}.[81,82]

Cornea consumes 38-90 µg/h of glucose (40-66% by epithelium) which is obtained by aqueous humor (less than 10% from limbal blood vessels and tears). This glucose is transferred to adenosine tri-phosphate (ATP) either by anaerobic pathway (Glycolytic pathway that brings the lactic acid and 2ATP) or by aerobic pathway [Tricarboxylic acid (TCA) cycle, in mitochondria-rich basal layer of epithelium which brings CO_2 and O_2 besides 36ATP]. TCA cycle occurs only in 15% of total glucose, so it obtains only 3 times more energy than anaerobic pathway. Due to hypoxia, glucose metabolism occurs by glycolytic anaerobic methods and lactic acid is produced. Lactic acid cannot be metabolized by cornea so it is diffused into the aqueous humour. Accumulation of lactic acid causes epithelial (satler's veil) and stromal oedema.

2.2.2 Conjunctiva

Conjunctiva is a vascular loose connective tissue that is present as a lining of the globe beyond the cornea, over the upper and lower fornices, under the upper and lower eyelids and at the limbus. It is a transparent mucous membrane. Bulbar conjunctiva covers the sclera and limbus.

Conjunctiva is made up of two layers: epithelium and stroma. Epithelium consists of 4 - 12 layers depending upon the location. Surface cells have microplicae and microvilli and it

contains basement membrane, but the basement membrane is not well organized.[83] Conjunctival stroma is made up of loosely but parallelly arranged collagen bundles. Numerous fibroblasts (main cell type) are present. It also contains many immunological cells like mast cells, macrophages, polymorphonuclear leucocytes, eosinophils and lymphocytes. Vascular supply of the conjunctiva is done by palpebral branches of the nasal and lacrimal arteries and also by anterior ciliary arteries.

Conjunctiva is rich of glands which are the sources of tears. Goblet cells are unicellular and sero-mucous secreting glands found in the epithelial layer of conjunctiva. These cells are absent in limbus and lid margins while found highly in bulbar region. Goblet cells have single-discharge life-cycle which secrete mucin. Glands of Wolfring are present at the upper limit of tarsal plate in conjunctival stroma and their structure and function are similar with the lacrimal gland. Glands of Krause are present in conjunctival stroma near fornices, 20 in the upper and 8 in the lower conjunctiva, more numerous laterally.[84] Similar in structure and function of the lacrimal gland, they contribute aqueous layer of the tears. Crypts of Henle are mucous crypts which are found in the superior peripheral palpebral conjunctiva.

2.2.3 Limbus

Limbus is a transition zone of the epithelium and connective tissues of the cornea, and the conjunctiva and sclera combined. It consists of 10-12 layers of epithelial cells unlike 5-6 layers of corneal epithelium.[83] These tissues do not have goblet cells but have melanocytes and blood vessels; so it is different from conjunctiva and cornea. Limbus is supplied by terminal arteries (which form peripheral cornea arcades) and recurrent arteries. Limbus is innervated by intrascleral and conjunctival nerves.

2.2.4 Lacrimal glands

Lacrimal glands are located in the lacrimal fossa of supero-temporal orbit. It is divided by *Levator Palpebral Superioris* muscle into a larger upper orbital portion (with 2-5 ducts) and a smaller lower palpebral portion (with 6-8 ducts). Lacrimal ducts open into superior palpebral conjunctiva.

Lacrimal gland is innervated by the lacrimal branch of ophthalmic division of trigeminal sensory nerve (afferent), facial nerve (efferent) and superior cervical ganglion and carotid plexus (sympathetic). Parasympathetic innervation by sphenopalatine ganglion is still not clear.

2.2.5 Eyelids

Eyelids are modified folds of skin which help in spreading tears and protect the eyes from foreign body and unnecessary light. It has four structural layers: skin, muscular layer, fibrous layer and the mucosal layer. Skin of the eyelid is loose and elastic cutaneous layer. It is rich in blood vessels, lymphatics and nerves. Muscular layer consists oval, concentric orbicularis oculi muscle. It also contains smooth Muller's muscle that helps the *levator palpebrae superioris* keep the eye open when awake. Fibrous layer is composed of fibrous and elastic tissues. It is made up of tarsal plate, orbital septum and Meibomian glands. The innermost mucosal layer is the palpebral conjunctiva.

Orbicularis oculi muscle contraction makes the zipper-like closure of eyelids from temporal to nasal. It is supplied by the facial nerve. Contraction of *levator palpebrae superioris* muscle (a little help from sympathetically innervated smooth *Muller's* muscle) opens the eyelids. It is innervated by oculomotor nerve. During blinking, lower lid almost does not move. Spontaneous blinking is usually a response to corneal dryness, irritation, anxiety, sound and air pollution.

Meibomian glands which are found in the upper lid (25 longer) and in the lower lid (20 shorter) are sebaceous glands and supply lipid of the tears.[67] This oily secretion helps to prevent the tear overflow. Glands of Zeis are sebaceous glands associated with lash follicles and supply lipid of the tears. Moll's Glands are modified sweat glands which open into the Zeis Glands.

2.2.6 Tear

Tear has an important role in ocular surface physiology. It maintains a smooth optical surface of the cornea and also maintains a moist environment for the epithelium of the cornea, conjunctiva and lids. Its bactericidal/bacteriostatic properties protect from infection. It is one of the media of nutrition support for the anterior cornea.

About half of the tear (4 µL) is found in cul-de-sac while some portion (1µL) is present on pre-corneal and as tear meniscus (3 µL).[85] The regular tear production, wetting the globe and drainage is aided by capillary action, gravity and blinking. Tear distribution is maintained by eyelid action and globe movement during blink. Lid movement spread the tears over the globe. Mucin layer enhances the wettability of epithelium. Lipid layer spreading over surface increases tear film thickness and stability. Tear turnover occurs about 16% per minute.

Tear flow is possible due to contraction of the *orbicularis oculi* muscle which causes lid closure with a scissor-like action towards the nose that propels the tears towards the medial canthus. Same time, lacrimal sac distends causing negative pressure which causes tear draw. Gravity and capillary action help in tear drainage. Tears from the upper and lower puncta flow to the nasal cavity through two canaliculi and nasolacrimal duct. The valve of Hasner guards the naso-lacrimal duct which prevents the reflux of the tear.

Tear structure

Basically, tear consists of three layered structure.[86] Innermost mucus layer, middle aqueous layer and outermost lipid layer. Mucus layer is 0.02-0.05 µm thick hydrophilic layer which enhances epithelial wettability. Adhesion of mucus layer is possible due to the presence of microvilli and microplicae in the superficial epithelial cells. Mucus is secreted by goblet cells and some from lacrimal glands. It is responsible for maintaining the stability of the tear film. Each blink resurfaces the epithelium with a renewed mucus layer. Aqueous layer is the thickest portion of the tears which is approximately 7 µm thick. It is secreted by lacrimal glands and in small amount by glands of Krause and Wolfrings. It is the only layer involved in true tear flow. It is the medium of oxygen and carbon dioxide transfer. Lipid Layer is the outermost thin film with thickness 0.1 µm. It is secreted by Meibomian glands and a small amount by Zeis gland. It protects the tears from evaporation. On blinking, it does not move, it just compressed and thickened.

Tear consists in 98.2 % of water. Normal osmolarity range is 294-334 mOsm/l with an average of 310 mOsm/l. Osmolarity decreases with eye closure by reduced evaporation. Reflex tearing by initial hard lens wear causes a reduction in osmolarity and initial soft lens wear increases tear evaporation due to blink rate increasing osmolarity. These alterations return to normal values within 1 week in hard and 2-3 days in SCL wearers.

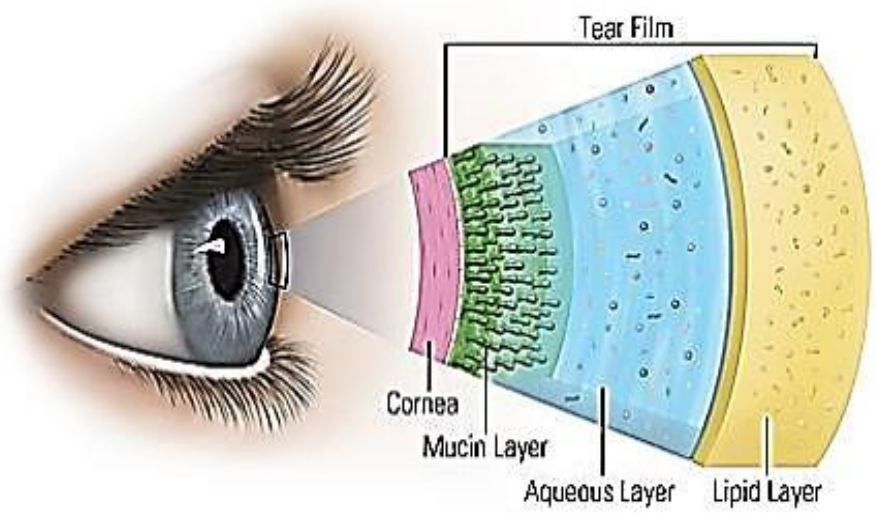

Figure 2.7. Tear film structure. [Figure obtained from http://www.lamellar.com/research-development/dry-eye-disease-ded/ assessed on October 24, 2015]

Tear volume is about 6.5 – 8 µL with flow rate 0.6 µL/min and turnover rate 16 % per minute. About 1-15 g tear is produced every day. Oxygen tension on tear surface is 155 mmHg during open eye and 55 mmHg during closed eye condition. It contains bactericidal/bacteriostatic proteins like lysozyme, lactoferrin and beta-lysin. Tear contains albumin, prealbumin, lysozyme, lactoferrin, transferrin and immunoglobulins.

Stimulation for the tear secretion may be psychogenic due to thought, emotion and sensory via the sensory root of the trigeminal nerve. It can be divided into basal (normal tears) and reflex (due to emotion, thought and injury).

Tear tests

Entire epithelium is coated by a loose carbohydrate called glycocalyx which binds tear mucus with epithelium. Cornea wets if $ST_{\text{tear film}} < ST_{\text{epithelium}} + ST_{\text{tear/epithelium interface}}$. Mucus makes ST of interface low. Narrow palpebral fissure width lowers ST of tears. Tear film break-up time (TBUT) can be measured with slit-lamp which is due to evaporation followed by localized thinning. Fluorescein dye with blue light and yellow barrier helps to easily detect the tear break-up. Less than 10 secs of TBUT is considered as abnormal. Non-invasive tear break-up time (NITBUT) is used with a grid or image projected onto the cornea and quality of the reflected image is assessed using various techniques.[87] It is more consistent and reliable than TBUT, but it is more

difficult because specular reflection is used in a narrow field. Schirmer's test is a cheaper and easier procedure to measure the aqueous portion of tears. A watman#41 filter paper strip is bent and inserted into the lower fornix (Figure 2.8). The wet length of the paper indicates the dry eye condition. Fluorophotometry is used to measure the tear flow rates. Phenol-red thread test is another test used to measure the basal tear volume.[88] The principle is similar with Schirmer's test but is more comfortable and faster. Interferometry technique is used to measure the overall thickness of the tear film and also the lipid layer thickness.[89]

Figure 2.8 Schirmer's test in a subject. [Image used with written permission of the subject].

2.3 Effects of soft contact lens on ocular surface

2.3.1 Effects of contact lenses on cornea

Cornea is avascular so it should get oxygen from the atmosphere, limbus or from palpebral conjunctiva for its metabolic activities.[90,91] During CL wear, the cornea can obtain oxygen from the dissolving tears or through the CL. Because of the larger diameter of the soft CL, cornea gets a negligible amount of oxygen by tears because only 1-2% of tears exchange during each blink.[92,93] So, the main source of oxygen for the anterior cornea during lens wear is from the Dk/t of the lens, however, CL wear acts as a barrier to oxygen influx and carbon dioxide efflux.[94]

Low Dk/t CL may cause different side effects on the ocular surface like corneal oedema, corneal microcysts, endothelial polymegethism and blebs.[91] Cornea shifts to acidic due to retention

of carbon dioxide. Corneal oedema occurs due to a reduction in the endothelial pump, retention of fluid and swelling due to the accumulation of lactic acid. To avoid the corneal swelling by hypoxia, CL should have a minimum Dk/t of 24.1± 2.7 X 10^{-9} barrer/cm or more i.e. on the corneal surface there should be equivalent oxygen percentage (EOP) of 9.9% when CL are worn on open eye condition.[77] But during overnight wear (CL wear on eye closure), CL should have a minimum Dk/t of 87.0 3.3X 10^{-9} barrer/cm (17.9% of EOP) which causes 4% of overnight corneal swelling equivalent to corneal swelling in non-lens wearers.[77] Harvitt and Bonanno found that 23 and 89 unit of Dk/t are required to avoid corneal swelling in open and closed eye lens wear respectively.[95] Later, they have suggested that to prevent corneal anoxia Dk/t of at least 35 units for open eye and 125 units for the closed eye condition are required.[96] Recently Papas suggested that Dk/t necessary to avoid corneal swelling is extremely different between central and peripheral cornea.[19] For a typical minus CL with central and peripheral thickness 80 and 220 µm respectively, the minimum required Dk is 20 barrer and 200 barrer for central and peripheral region, respectively. During sleep *i.e*. closed-eye condition, oxygen tension decreases to 55 mmHg. In this condition, overnight sleep i.e. 8 hours sleep causes 3% to 5.5% corneal swelling which resolves quickly during eye opening after awake.[97,98]

Fitting of SCL also affects the corneal swelling. A recent study found that tight fitting produces more swelling of the peripheral cornea than that of loose fitting.[99] The reason behind this has not been clear, but it may be due to more tear exchange under the loose fitting SCL.

Chronic hypoxic stress reduces corneal sensitivity,[100] increases epithelial fragility,[90] compromises epithelial adhesion,[101,102] produces epithelial microcysts,[103] affecting all corneal layers. Epithelial microcysts are the accumulations of metabolic byproducts, cellular debris or necrotic tissue due to change in metabolic activities of the cornea which appear as small round inclusions within epithelium in CL wearers.[74,104,105] Microcysts are rarely found with high Dk/t lenses.[106]

Animal studies show that CL wear retard the corneal epithelial cell proliferation rate[107,108] which supports that lens wear retard the mitosis of the epithelial cells.[109] High Dk/t lenses affect less on epithelial cell mitosis and the proliferation rate.[107] Epithelial thinning has been found in

long-term extended wearers.[74] The higher the Dk/t, the less is the epithelial thinning.[110,111] This may be due to the alteration of epithelial cell proliferation or exfoliation by lens wear.

CL wear reduces epithelial permeability[112] and this effect is higher in long-term lens wearers; however, it may not be related with lens related hypoxia.[113] A current study showed that continuous wear of SiHy lenses for 30 days increases epithelial permeability.[114]

Corneal staining is one of the indicators of corneal health and its strength shows the severity of the corneal problem. Corneal staining is found common in SCL wearers and may be due to lens material characteristics, lens design and lens fittings.[115] It was found associated with lens and lens care product as well as their combinations.[116] Due to incomplete blinking or lid closure effect, corneal staining was found different on different regions.[117-119] Inferior cornea is more susceptible on CL induced staining[120] and is also dependent upon lens power.[120]

Stroma

Stromal swelling is a well-established side effect of SCL wear specifically with extended wear of low Dk/t lenses.[19] It is due to the imbalance in corneal biophysico-chemical properties of the cornea.[121] Hypoxia reduces aerobic metabolism and increases anaerobic glycolysis which accumulates lactate in the stroma increasing acidosis. This changes osmotic gradient sufficiently that endothelial pump can no longer maintain equilibrium and results net inflow of fluid into stroma which causes swelling.[19] Swelling is found inversely proportional to the Dk/t of the lenses.[77,121] Hypoxia also increases touch threshold.[122] It is estimated that a minimum of 8% EOP is necessary to maintain corneal sensitivity.[123] Studies have shown thinning of stroma with long-term extended wear of low Dk lenses.[74] This may be due to loss of glycosaminoglycans by leakage during stromal oedema.[124] Moreover, CL wear reduces the stromal keratocyte density which may not be related with Dk/t of the lenses.[125]

Corneal vascularization occurs due to engorgement of limbal blood vessels into cornea or by development of new vessels in the cornea.[19] It is common in SCL wear specifically with extended wear modality[126] but is hardly found with high Dk/t lenses.[127]

Endothelium

Closed eye or lens wear or hypoxia reduces pH of cornea [makes more acidic][128] which causes transient change in endothelial cell mosaic called endothelial bleb response.[129,130] To prevent pH change, at least 18 Dk/t *i.e.* 8% EOP is necessary which is very less in comparison to the Dk/t required to maintain pH in corneal epithelium and stroma.[131] Morphological changes in both shape and size of endothelial cells are observed in CL wearers especially in low Dk/t lens in extended modality.[132] Endothelial cell density also decreases with long-term CL wear.[132]

Corneal biomechanical properties

Due to placement of CL directly over the cornea, corneal biomechanical properties may be affected by CL wear. Effect of CL on corneal biomechanical properties has not been known in normal condition. But, it is found that corneal oedema induced by low Dk/t lenses on closed eye condition increases corneal resistance factor (CRF) but does not change corneal hysteresis (CH).[133] So, it is considered that CH is relatively stable with CL wear in comparison to CRF. The findings of Franco and Lira also supports this fact that CRF highly correlates with the corneal thickness but CH does not.[134] This indicates that CL with Dk/t values more than 24 units do not induce corneal swelling if worn on daily wear modality[77] and thus do not change the corneal biomechanical properties. Chen *et al*. found a decrease in CRF with short-term orthokeratology lens wear.[135] Although they have suggested that corneal shape change may have some role on CRF, latter studies show that there is no role of anterior corneal curvature on corneal biomechanical properties.[134] Corneal reshaping therapy (CRT) lens wear reduces CH but does not change CRF.[136] However, these effects of change on corneal biomechanical properties may be reversible, once the lenses are not worn, corneal biomechanical properties may return back to normal level.[136]

Corneal sensitivity

Studies found that corneal sensitivity decreases with SCL wear.[100] This effect is minimum with silicone lenses.[137] CL wear may reduce corneal sensitivity due to an effect of hypoxia[123] or other mechanical effects of lenses.[138] Some studies suggest that long-term wear of CL returns the corneal sensitivity to normal.[137] However, conjunctival sensitivity is found increased with daily wear SiHy lenses.[139]

2.3.2 Effects of contact lenses on limbus

Limbal redness is common in SCL wearers.[19] It is the most prevalent sign of CL induced hypoxia which can be detected as early as 4 hours of lens insertion.[140,141] Because it supplies the blood to the peripheral cornea, when cornea feels hypoxia, limbal blood flow rapidly increases causing limbal hyperemia.[142] Even short-term high Dk/t lenses induce some limbal redness.[143,144] Little change in limbal hyperemia with hyper Dk/t CL indicates that CL wear induced limbal hyperemia is due to hypoxia and can be eliminated with high Dk/t SiHy lenses.[141] Studies show that Dk/t of more than 36 barrer/cm does not produce excessive redness during daily wear modality[143] but to maintain the exactly same state of redness similar to non-CL wearers, Dk/t of about 125 barrers/cm is necessary on open eye condition.[142] Changes on limbal physiology due to CL wear is found to be related with lens material and wearing modality.[145] Moreover, studies show that lens, lens care products and their combination affect limbal redness.[116] Same lens material may induce a different level of limbal redness with different lens care products. Exposed part of the limbus that is nasal and temporal limbus may be affected higher than other non-exposed parts due to a difference in evaporation of the tears.[146] Long-term limbal redness can facilitate corneal neovascularization.[147] Limbal stem cell density is found to be decreased with CL wear which may be due to hypoxia related complication.[148]

2.3.3 Effect of CL on conjunctiva

Dilution of blood vessels in conjunctiva makes it hyperemic or redness and further chemosis of the tissues around the blood vessels makes it pinkish.[149] Conjunctival hyperemia is higher in CL wearers.[143] The cause of redness in conventional CL lens wearers may be hypoxia.[150] However, an increase in bulbar redness in short-term modern high Dk/t CL wearers was also found in latest studies.[144,143] Lens deposits,[151] mechanical irritation, improper lens fittings,[115] lens edge profile, modulus, solution toxicity or lens mediated conjunctival inflammatory events[152] may also contribute to conjunctival redness. Due to evaporation of tears on exposed parts of the bulbar conjunctiva – temporal and nasal part – conjunctival redness is higher in comparison to the superior and inferior bulbar conjunctiva.[146]

Conjunctival staining can be found in CL wearers, in limbus or CL edge or in bulbar and palpebral area even after a short time of lens wear.[143] Thin post lens tear film in the peripheral lens

region may cause conjunctival staining on bulbar conjunctiva around limbus. Lens material characteristics, lens designs and lens fittings may also be related with conjunctival staining. This can be observed clearly with lissamine green which stains by entering membrane-compromised epithelial cells.[153] Fluorescein which stains the ocular epithelial surface with compromised cells,[154] is also used to examine the conjunctival staining. There are inconsistent reports on the effects of conjunctival staining on CL comfort.[155,156] Improper lens fit, especially steeper fit or lenses with higher rigidity can make a depression on conjunctiva due to the pressure of the edge of the lens.[157] These conjunctival indentations are seen as fluorescein pooling on sodium fluorescein dye. Conjunctival staining less than grade 2 in Efron grading is considered as clinically non-significant.[156]

Physical movement of lenses may cause delamination and bunching-up of the conjunctival layers creating conjunctival flaps.[158] It is found higher in lenses with higher-modulus[159,160] and with a sharp-edge lens.[160] Similarly, such incidences were found higher in extended wear modality of SiHy lenses[160,161] in comparison to the daily wear modality.[160,161] The potential risk of conjunctival flaps is still unclear.[158] A round-edged lens with lower modulus may reduce this effect of CL wear.[162] Lid parallel conjunctival folds (LPCOFs) are also common in soft CL wearers, especially with SiHy lenses.[162] Its etiology and the consequence are still unknown, but its positive correlation with fluorescein staining, tear break-up time may indicate that LPCOFs are related to CL induced dry eye.[163]

The cells of the corneal epithelium are constantly regenerating and migrating anteriorly where they become desquamated and shed out into the tear film. Stem cells which are found in limbal epithelium demonstrate unlimited self-renewal which helps in constant regeneration and migration of corneal epithelial cells.[164] They also protect the migration of conjunctival cells towards cornea.[165] CL wear causes stem cell deficiency which may be due to lens-induced hypoxia or mechanical friction.[165] Early stage of stem cell deficiency may be asymptomatic but with progression, it may enhance corneal inflammation and discomfort.[165] In early cases, just cease of lens wear and application of lubricants can sooth the problem but in latter stage, stem cell transplant is necessary.[166]

Conjunctival cytology

Previous studies conducted in conjunctival cytology have compared GCD and epithelial cell morphology with CL wearers and non-CL wearers.[167-169] Research found a decrease in mucin level in CL wearers in comparison to non-CL wearers.[170,171] Effect of CL wear on conjunctival cytology may be due to cytotoxic effects attributable to CL wear and preservatives and chelating agents in SCL care system.[172] Studies show contradictory reports on the effect of CL wear on GCD. It is because GCD is highly variable from one person to another even in normal condition.[167] Most of the previous studies show a decrease in GCD due to CL wear.[167,173,174] However, a few studies found an increase in GCD after SCL wear which may be due to adaptive response to ocular surface.[175,176]

Previous studies found higher epithelial metaplasia in subjects who wear SCL in comparison to non-lens wearers.[167,173,174,177-180] Conjunctival epithelial cell metaplasia and GCD change can be found in short-term lens wearers while snake like chromatin in epithelial cell nucleus can be found in long-term CL wearers.[172]

2.3.4 Other effects of contact lenses

Refractive error

Daily wear of SCL does not significantly change the refractive error. But, some recent studies show that long-term extended wear of SCL can change some refractive error. A recent study found that low Dk/t lenses increase myopia progression by 0.3D however, high Dk/t SiHy lenses do not have any impact on refractive error during nine months of extended wear.[181] Progression of myopia due to low Dk/t lenses may be due to its hypoxia effect and no change in myopia with SiHy lenses may be due to physical pressure on the anterior surface of the cornea.[182] Dubmleton *et al*. found flattening of the anterior corneal surface by extended wear SiHy lenses.[181] However, later they found that change in corneal curvature does not correlate with Dk/t.[183]

Intraocular pressure

There are controversial reports about the accuracy of intraocular pressure (IOP) measurement over soft CL.[184-195] Majority of the studies concluded that CL should be removed for the accurate measurement of IOP although the measurement may not be clinically significantly different with the IOP values without lenses. Moreover, accuracy depends upon the lens material,

lens design as well as the measuring instruments. The relation between the corneal thickness and IOP is well established – the higher the corneal thickness, the higher the IOP and vice-versa.[196,197] If CL wear induces corneal swelling, it increases the value of IOP and if CL wear makes the cornea thinner, IOP might be lower. Short time SCL wear was found to reduce the IOP.[198,199] However, these studies were conducted with CL wear for just 2 hours. So, the effect of long-term CL wear on IOP has not still been established.

Contrast sensitivity

Many studies found that normal SCL wear reduces the visual contrast sensitivity (VCS).[200-202] Changes in corneal physiology like corneal oedema due to CL wear may be a cause of VCS reduction.[203-205] However, specially designed CL can improve VCS for example, sport-tinted CL.[206] Similarly, filter CL were found effective to improve VCS in subjects with retinitis pigmentosa.[207] Some studies show that VCS is better with SCL in comparison to spectacles.[208] The difference in VCS depends upon lens materials, designs, thickness and manufacturing process.[209,210] However some of the studies did not get any changes on VCS with SCL wear.[204,210]

References

1. Hofstetter H. Leonardo's contact concept. *CL Forum* 1984;9:15-17.

2. Heitz R. Leonardo da Vinci did not invent contact lenses. *CLAO J.* 1983;9(4):313-316.

3. Fatt I. Sir John Fredrick William Herschel: The man and his time. *Optician* 1993;206:26-27.

4. Heitz R. History of Contact Lenses. In: Dabezies OH, Contact Lenses: The CLAO Guide to Basic Science and Clinical Practice, Vol. 1, Update 3. Grune & Stratton Inc, Orlando. 1.1 - 2.3.; 1984.

5. Efron N, Pearson RM. Centenary celebration of Fick's Eine Contactbrille. *Arch. Ophthalmol.* 1988;106(10):1370-1377.

6. Nissel G. The Müllers of Wiesbaden. *Cont Len J* 1977;11:15-19.

7. Refojoo M, Dabezies O. Classifications of the Types of Materials Used for Contact Lenses. In: Dabezies OH, Contact Lenses: The CLAO Guide to Basic Science and Clinical Practice, Vol. 1, Update 3. Grune & Stratton Inc, Orlando. 11.2.; 1984.

8. Knoll H. William Feinbloom: Pioneer in plastic contacts. *CL Forum* 1977;1(8):29-32.

9. Bailey N. The contact lens - past, present and future. *CL Spectr.* 1987;2:6-51.

10. Mandell R. Contact Lens Practice. 4th Ed. Charles C. Thomas, Springfield. 14-15.; 1988.

11. Marking a Milestone in Contact Lenses. *CL Spectr.* 1996;(March):assessed on 13 September 2015.

12. Stahl N, Reich L, Ivani E. Report on laboratory studies and preliminary clinical application of a gas permeable plastic contact lens. *J Am Optom* 1974;45:302-307.

13. Fonn D, Dumbleton K, Jalbert I, Sivak A. Benefits of Silicone Hydrogel Lenses. *Contact Lens Spectr.* 2006;21(Suppl):38-44.

14. Tighe BJ. A Decade of Silicone Hydrogel Development: Surface Properties, Mechanical Properties, and Ocular Compatibility. *Eye Contact Lens* 2013;39(1):4-12.

15. Fatt I. Oxygen Transmission. In: Bennett ES, Weissman BA. Clinical Contact Lens Practice. JB Lippincott Company, Philadelphia.; 1992.

16. Alvord L, Court J, Davis T, *et al*. Oxygen permeability of a new type of high Dk soft contact lens material. *Optom. Vis. Sci.* 1998;75(1):30-36.

17. Winterton L, White J, Su K. Cuolometric method for measuring oxygen flux and Dk of contact lenses and lens materials. *ICLC* 1987;14:441-52.

18. Fatt I, St Helen R. Oxygen tension under an oxygen-permeable contact lens. *Am. J. Optom. Arch. Am. Acad. Optom.* 1971;48(7):545-555.

19. Papas EB. The significance of oxygen during contact lens wear. *Contact Lens Anterior Eye* 2014;37(6):394-404.

20. Mirejovsky D, Patel AS, Young G. Water properties of hydrogel contact lens materials: A possible predictive model for corneal desiccation staining. *Biomaterials* 1993;14(14):1080-1088.

21. Holden BA, Sweeney DF, Seger RG. Epithelial erosions caused by thin high water content lenses. *Clin. Exp. Optom.* 1986;69(3):103-107.

22. Szczotka-Flynn L. Lens Distinctions. *Contact Lens Spectr.* 2007;22(June).

23. Jacob JT. Biocompatibility in the development of silicone-hydrogel lenses. *Eye Contact Lens* 2013;39(1):13-9.

24. Nicolson PC, Vogt J. Soft contact lens polymers: An evolution. *Biomaterials* 2001;22(24):3273-3283.

25. Efron N, Morgan PB, Cameron ID, Brennan NA, Goodwin M. Oxygen permeability and water content of silicone hydrogel contact lens materials. *Optom. Vis. Sci.* 2007;84(4):328-337.

26. Hui A. More Than Just Juice Updates in Lens Care. *CL Spectr.* 2015;30:30-33.

27. Hutter JC, Green JA, Eydelman MB. Proposed Silicone Hydrogel Contact Lens Grouping System for Lens Care Product Compatibility Testing. *Eye Contact Lens* 2012;38(6):358-362.

28. Maldonado-Codina C, Morgan PB. In vitro water wettability of silicone hydrogel contact lenses determined using the sessile drop and captive bubble techniques. *J. Biomed. Mater. Res. - Part A* 2007;83(2):496-502.

29. Guillon M, McGrogan L, Guillon JP, Styles E, Maissa C. Effect of material ionicity on the performance of daily disposable contact lenses. *Contact lens anterior eye* 1997;20(1):3-8.

30. Keir N, Jones LW. Wettability and Silicone Hydrogel Lenses : A Review. *Eye Contact Lens* 2013;39(1):100-108.

31. Giordano GG, Refojo MF. Polymer Chemistry. In Kastl PR, Ed. Contact Lenses The CLAO Guide to Basic Science and Clinical Practice. Volume I Basic Science. Dubuque, IA KendallHunt Publishing Company; 1995.

32. Bron A, Tiffany J, Gouveia S, Yokoi N, Voon L. Functional aspects of the tear film lipid layer. *Exp. Eye Res.* 2004;78(3):347-60.

33. Grobe G, Valint P, Ammon D. Surface chemical structure for soft contact lenses as a function of polymer processing. *J Biomed Mater Res* 1996;32(1):45-54.

34. Holly FJ. Tear film physiology and contact lens wear. II. Contact lens-tear film interaction. *Am. J. Optom. Physiol. Opt.* 1981;58(4):331-341.

35. Nichols JJ, King-Smith PE. The impact of hydrogel lens settling on the thickness of the tears and contact lens. *Investig. Ophthalmol. Vis. Sci.* 2004;45(8):2549-2554.

36. Nichols JJ, Mitchell GL, King-Smith PE. Thinning rate of the precorneal and prelens tear films. *Investig. Ophthalmol. Vis. Sci.* 2005;46(7):2353-2361.

37. Sarac O, Gurdal C, Bostanci-Ceran B, Can I. Comparison of tear osmolarity and ocular comfort between daily disposable contact lenses: Hilafilcon B hydrogel versus narafilcon A silicone hydrogel. *Int. Ophthalmol.* 2012;32(3):229-233.

38. Miller WL, Doughty MJ, Narayanan S, *et al*. A comparison of tear volume (by tear meniscus height and phenol red thread test) and tear fluid osmolality measures in non-lens wearers and in contact lens wearers. *Eye Contact Lens* 2004;30(3):132-137..

39. Panaser A, Tighe BJ. Function of lipids - their fate in contact lens wear: An interpretive review. *Contact Lens Anterior Eye* 2012;35(3):100-111.

40. Dalton K, Subbaraman L. Physical Properties of Soft Contact Lens Solutions. *Optom. Vis. Sci.* 2008;85(2):122-128.

41. Ketelson HA, Perry S, Sawyer G. Exploring the Science and Technology of Contact Lens Comfort. *Contact Lens Spectr.* 2011;(September).

42. Fatt I. Prentice Medal lecture contact lens wettability--myths, mysteries, and realities. *Am. J. Optom. Physiol. Opt.* 1984;61(4):419-30.

43. Holly F, Refojo M. Wettability of hydrogels I Poly(2-hydroxyethyl methacrylate). *J Biomed Mater Res* 1975;9(3):315-26.

44. Meslin D, Obrecht G. Effect of chromatic dispersion of a lens on visual acuity. *Am. J. Optom. Physiol. Opt.* 1988;65(1):25-28.

45. Papas EB, Carnt N, Willcox MDP, Holden BA. Complications associated with care product use during silicone daily wear of hydrogel contact lens. *Eye Contact Lens* 2007;33(6 Pt 2):392-393; discussion 399-400.

46. Szczotka-Flynn L, Diaz M. Risk of corneal inflammatory events with silicone hydrogel and low dk hydrogel extended contact lens wear: a meta-analysis. *Optom. Vis. Sci.* 2007;84(4):247-256.

47. Jones L, Evans K, Sariri R, Franklin V, Tighe B. Lipid and protein deposition of N-vinyl pyrrolidone-containing group II and group IV frequent replacement contact lenses. *CLAO J* 1997;23(2):122-126.

48. Hough T. Shedding light on a new high-volume contact lens manufacturing process. *Contact Lens Spectr.* 1998;13(March):42-44.

49. Sweeney DF, Holden BA. Silicone elastomer lens wear induces less overnight corneal edema than sleep without lens wear. *Curr. Eye Res.* 1987;6(12):1391-1394.

50. Chou BB. The Evolution of Silicone Hydrogel Lenses. *Contact Lens Spectr.* 2008;23(June).

51. Tighe B, Brennan N, Coles C. Silicone hydrogels - what are they and how should they be used in everyday practice? *Optician* 1999;218(5726):31-32.

52. Marsden HJ, Geffen DI, Giedd KA, Jedlicka J, Wesley G. A New Way to See the World. *Contact Lens Spectr.* 2013;28(3):Supplement.

53. Jones L, Senchyna M. Soft Contact Lens Solutions Review Part 1: Components of Modern Care Regimens. Optometry in Practice 2007;8:45-56.

54. Efron N, Morgan PB, Woods CA. Trends in Australian contact lens prescribing during the first decade of the 21st Century (2000-2009). *Clin. Exp. Optom.* 2010;93(4):243-252.

55. Efron N, Morgan PB. Soft contact lens care regimens in the UK. *Contact Lens Anterior Eye* 2008;31(6):283-284.

56. Willcox MDP. Solutions for Care of Silicone Hydrogel Lenses.pdf. *Eye Contact Lens* 2013;39(1):24-8..

57. Nikolic M, Kilvington S, Cheung S, *et al*. Survival and growth of Stenotrophomonas maltophilia in multipurpose contact lens solutions. Invest Ophthalmol Vis Sci 2010;51:E-Abstract 1540.

58. Jones L, Powell CH. Uptake and release phenomena in contact lens care by silicone hydrogel lenses. *Eye Contact Lens* 2013;39(1):29-36.

59. Broxton P, Woodcock PM, Gilbert P. A study of the antibacterial activity of some polyhexamethylene biguanides towards Escherichia coli ATCC 8739. *J. Appl. Bacteriol.* 1983;54(3):345-353.

60. Codling CE, Maillard JY, Russell a. D. Aspects fo the antimicrobial mechanisms of action of a polyquaternium and an amidoamine. *J. Antimicrob. Chemother*. 2003;51(5):1153-1158.

61. Gromacki S, Ward M. Understanding Contemporary Contact Lens Care Products. *Contact Lens Spectr.* 2013;28:20-25.

62. Hogan M, Avarado J, JE W. Histology of the Human Eye. W.B. Saunders Company, Philadelphia.; 1971.

63. DelMonte DW, Terry K. Anatomy and physiology of the cornea. *J Cataract Refract Surg* 2011;37:588-98.

64. Huang J, Ding X, Savini G, *et al*. Central and Midperipheral Corneal Thickness Measured with Scheimpflug Imaging and Optical Coherence Tomography. *PLoS One* 2014;9(5):e98316.

65. Patel S V., McLaren JW, Hodge DO, Bourne WM. Confocal microscopy in vivo in corneas of long-term contact lens wearers. *Investig. Ophthalmol. Vis. Sci.* 2002;43(4):995-1003.

66. Ameen DB, Bishop MF, McMullen T. A lattice model for computing the transmissivity of the cornea and sclera. *Biophys. J.* 1998;75(5):2520-2531.

67. Warwick R. Eugene Wolff's Anatomy of the Eye and Orbit. 7th Ed. H.K. Lewis & Co. Ltd, London.; 1976.

68. Reinstein DZ, Archer TJ, Gobbe M, Silverman RH, Coleman DJ. Epithelial thickness in the normal cornea: three-dimensional display with very high frequency ultrasound. *J. Refract. Surg.* 2008;24(6):571-581.

69. Jalbert I, Sweeney DF, Stapleton F. The effect of long-term wear of soft lenses of low and high oxygen transmissibility on the corneal epithelium. *Eye* 2009;23(6):1282-7.

70. Cenedella RJ, Fleschner CR. Kinetics of corneal epithelium turnover in vivo: Studies of lovastatin. *Investig. Ophthalmol. Vis. Sci.* 1990;31(10):1957-1962.

71. McCaa CS. The eye and visual nervous system: anatomy, physiology and toxicology. *Environ. Health Perspect.* 1982;Vol. 44:1-8.

72. Fini ME, Stramer BM. How the cornea heals: cornea-specific repair mechanisms affecting surgical outcomes. *Cornea* 2005;24(8 Suppl):S2-S11..

73. Nishida T, Yasumoto K, Otori T, Desaki J. The network structure of corneal fibroblasts in the rat as revealed by scanning electron microscopy. *Investig. Ophthalmol. Vis. Sci.* 1988;29(12):1887-1890.

74. Holden BA, Sweeney DF, Vannas A, Nilsson KT, Efron N. Effects of long-term extended contact lens wear on the human cornea. *Invest. Ophthalmol. Vis. Sci.* 1985;26(11):1489-1501.

75. Müller LJ, Pels E, Vrensen GF. The specific architecture of the anterior stroma accounts for maintenance of corneal curvature. *Br. J. Ophthalmol.* 2001;85(4):437-443.

76. Klyce SD. Enhancing fluid secretion by the corneal epithelium. *Investig. Ophthalmol. Vis. Sci.* 1977;16(10):968-973.

77. Holden BA, Mertz GW. Critical oxygen levels to avoid corneal edema for daily and extended wear contact lenses. *Invest. Ophthalmol. Vis. Sci.* 1984;25(10):1161-7.

78. Brennan NA. A model of oxygen flux through contact lenses. *Cornea* 2001;20(1):104-108.

79. Takatori S, de la Jara P, Holden B, Ehrmann K, Ho A, Radke C. In Vivo Oxygen Uptake into the Human Cornea. *Invest. Ophthalmol. Vis. Sci.* 2012;53(11):6828-6828.

80. Takatori SC, de la Jara PL, Holden B, Ehrmann K, Ho A, Radke CJ. In vivo corneal oxygen uptake during soft-contact-lens wear. *Investig. Ophthalmol. Vis. Sci.* 2013;54(5):3472-3478.

81. Brennan NA. Beyond flux: total corneal oxygen consumption as an index of corneal oxygenation during contact lens wear. *Optom. Vis. Sci.* 2005;82(6):467-472.

82. Bonanno JA, Stickel T, Nguyen T, *et al*. Estimation of human corneal oxygen consumption by noninvasive measurement of tear oxygen tension while wearing hydrogel lenses. *Investig. Ophthalmol. Vis. Sci.* 2002;43(2):371-376.

83. Feng Y, Simpson TL. Corneal, limbal, and conjunctival epithelial thickness from optical coherence tomography. *Optom. Vis. Sci.* 2008;85(9):E880-E883.

84. Steuhl K-P. Ultrastructure of the Conjunctival Epithelium. A Monograph in the Series: Developments in Ophthalmology. Straub W (Ed.). S Karger AG, Basel.; 1989.

85. Mahmood M, Farris R, Lemp M. Lacrimal Physiology and Contact Lens Wear. In: Dabezies OH (Ed.), Contact Lenses: the CLAO Guide to Basic Science and Clinical Practice. 3rd ed. Grune & Stratton, Inc., Orlando. In: ; 1984.

86. Smolin G, Thoft R. (Eds.) The Cornea: Scientific Foundations and Clinical Practice. 2nd Ed. Little, Brown and Company, Boston.; 1987.

87. Nichols J, Nichols K, Puent B, Saracino M, Mitchell G. Evaluation of tear film interference patterns and measures of tear break-up time. *Optom. Vis. Sci.* 2002;79(6):363-9.

88. Hamano H, Hori M, Hamano T, *et al*. A new method for measuring tears. *CLAO J.* 1983;9(3):281-289.

89. Dubra A, Paterson C, Dainty C. Study of the tear topography dynamics using a lateral shearing interferometer. *Opt Express* 2004;12(25):6278-88.

90. Morgan PB, Woods CA, Tranoudis IG, *et al*. International contact lens prescribing in 2011. *Contact lens Spectr*. 2012;27(January):26-32.

91. Sweeney DF. Have silicone hydrogel lenses eliminated hypoxia? *Eye Contact Lens* 2013;39(1):53-60.

92. Polse K. Tear flow under hydrogel contact lenses. *Investig. Ophthalmol. Vis. Sci.* 1979;18(4):409-413.

93. McNamara NA, Polse KA, Brand RJ, Graham AD, Chan JS, McKenney CD. Tear mixing under a soft contact lens: Effects of lens diameter. *Am. J. Ophthalmol.* 1999;127(6):659-665.

94. Ruben M, Guillon M. Contact Lens Practice. Chapman and Hall Medical, London.; 1994.

95. Harvitt DM, Bonanno JA. pH dependence of corneal oxygen consumption. *Investig. Ophthalmol. Vis. Sci.* 1998;39(13):2778-2781.

96. Harvitt DM, Bonanno JA. Re-evaluation of the oxygen diffusion model for predicting minimum contact lens Dk/t values needed to avoid corneal anoxia. *Optom. Vis. Sci.* 1999;76(10):712-719.

97. Sweeney D. Factors Contributing to the Human Corneal Oedema Response. PhD Thesis. School of Optometry, The University of New South Wales, Sydney. 1991.

98. Harper CL, Boulton ME, Bennett D, *et al*. Diurnal variations in human corneal thickness. *Br. J. Ophthalmol.* 1996;80(12):1068-1072.

99. Holden BA, McNally JJ, Egan P. Limited lateral spread of stromal edema in the human cornea fitted with a (’donut') contact lens with a large central aperture. *Curr. Eye Res.* 1988;7(6):601-605.

100. Murphy PJ, Patel S, Marshall J. The effect of long-term, daily contact lens wear on corneal sensitivity. *Cornea* 2001;20(3):264-269. doi:10.1097/00003226-200104000-00006.

101. Madigan MC, Holden BA, Kwok LS. Extended wear of contact lenses can compromise corneal epithelial adhesion. *Curr. Eye Res.* 1987;6(10):1257-1260.

102. Madigan MC, Holden BA. Reduced epithelial adhesion after extended contact lens wear correlates with reduced hemidesmosome density in cat cornea. *Investig. Ophthalmol. Vis. Sci.* 1992;33(2):314-323.

103. Humphreys JA, Larke JR, Parrish ST. Microepithelial cysts observed in extended contact-lens wearing subjects. *Br J Ophthalmol* 1980;64(12):888-889.

104. Brown N. Visibility of transparent objects in the eye by retroillumination. *Br. J. Ophthalmol.* 1971;55(8):517-524.

105. Holden BA, Sweeney DF. The significance of the microcyst response: a review. *Optom. Vis. Sci.* 1991;68(9):703-707.

106. Covey M, Sweeney DF, Terry R, Sankaridurg PR, Holden BA. Hypoxic effects on the anterior eye of high-Dk soft contact lens wearers are negligible. *Optom. Vis. Sci.* 2001;78(2):95-99.

107. Ladage PM, Ren DH, Petroll WM, Jester J V., Bergmanson JPG, Cavanagh HD. Effects of eyelid closure and disposable and silicone hydrogel extended contact lens wear on rabbit corneal epithelial proliferation. *Investig. Ophthalmol. Vis. Sci.* 2003;44(5):1843-1849.

108. Ladage PM, Yamamoto K, Ren DH, *et al*. Proliferation rate of rabbit corneal epithelium during overnight rigid contact lens wear. *Investig. Ophthalmol. Vis. Sci.* 2001;42(12):2804-2812.

109. Hamano H, Hori M, Hamano T, *et al*. Effects of contact lens wear on mitosis of corneal epithelium and lactate content in aqueous humor of rabbit. *Jpn. J. Ophthalmol.* 1983;27(3):451-458.

110. Ren DH, Yamamoto K, Ladage PM, *et al*. Adaptive effects of 30-night wear of hyper-O2 transmissible contact lenses on bacterial binding and corneal epithelium: A 1-year clinical trial. *Ophthalmology* 2002;109(1):27-39.

111. Pérez JG, Méijome JMG, Jalbert I, Sweeney DF, Erickson P. Corneal epithelial thinning profile induced by long-term wear of hydrogel lenses. *Cornea* 2003;22(4):304-7.

112. Lin MC, Soliman GN, Song MJ, *et al*. Soft contact lens extended wear affects corneal epithelial permeability: Hypoxic or mechanical etiology? *Contact Lens Anterior Eye* 2003;26(1):11-16.

113. McNamara NA, Chan JS, Han SC, Polse KA, McKenney CD. Effects of hypoxia on corneal epithelial permeability. *Am. J. Ophthalmol.* 1999;127(2):153-157.

114. Lin MC, Yeh TN, Graham AD, *et al*. Ocular surface health during 30-day continuous wear: Rigid gas-permeable versus silicone hydrogel hyper-O 2 transmitted contact lenses. *Investig. Ophthalmol. Vis. Sci.* 2011;52(6):3530-3538.

115. Young G, Coleman S. Poorly fitting soft lenses affect ocular integrity. *CLAO J* 2001;27(2):68-74.

116. Jones LW, Macdougall N, Sorbara GL. Asymptomatic Corneal Staining Associated with the Use of Balafilcon Silicone-Hydrogel Contact Lenses Disinfected with a Polyaminopropyl. *Optom. Vis. Sci.* 2002;79(12):753-761.

117. Carolyn G Begley, Joseph T Barr, Timothy B Edrington, William D Long, Curtis D McKenney RLC. Characteristics of Corneal Staining in Hydrogel lenses. *Optom. Vis. Sci.* 1996;73(3):193-200.

118. Schwallie J, Mckenney C, William L, Angela M. Corneal Staining Patterns in Normal Non Contact Lens Wearers.pdf. *Optom. Vis. Sci.* 1997;74:92-98.

119. Bandamwar KL, Garrett Q, Cheung D, *et al*. Onset time course of solution induced corneal staining. *Contact Lens Anterior Eye* 2010;33(4):199-201.

120. Nichols KK, Mitchell GL, Simon KM, Chivers D, Edrington TB. Corneal staining in hydrogel lens wearers. *Optom. Vis. Sci.* 2002;79(1):20-30.

121. Leung BK, Bonanno JA, Radke CJ. Oxygen-deficient metabolism and corneal edema. *Prog. Retin. Eye Res.* 2011;30(6):471-492.

122. Millodot M. A review of research on the sensitivity of the cornea. *Ophthalmic Physiol. Opt.* 1984;4(4):305-318.

123. Millodot M, O'Leary DJ. Effect of oxygen deprivation on corneal sensitivity. *Acta Ophthalmol.* 1980;58(3):434-439.

124. Kangas TA, Edelhauser HF, Twining SS, O'Brien WJ. Loss of stromal glycosaminoglycans during corneal edema. *Investig. Ophthalmol. Vis. Sci.* 1990;31(10):1994-2002.

125. Ohta K, Shimamura I, Shiraishi A, Ohashi Y. Confocal Microscopic Observations of Stromal Keratocytes in Soft and Rigid Contact Lens Wearers. *Cornea* 2012;31(1):66-73.

126. Keech P, Ichikawa L, Barlow W. A prospective study of contact lens complications in a managed care setting. *Optom. Vis. Sci.* 1996;73(10):653-8.

127. Fonn D, MacDonald KE, Richter D, Pritchard N. The ocular response to extended wear of a high Dk silicone hydrogel contact lens. *Clin. Exp. Optom.* 2002;85(3):176-182.

128. Bonanno JA, Polse KA. Corneal acidosis during contact lens wear: Effects of hypoxia and CO2. *Investig. Ophthalmol. Vis. Sci.* 1987;28(9):1514-1520.

129. Williams L. Transient Endothelial changes in the in vivo Human Cornea. PhD Thesis, The University of New South Wales, Sydney. 1986.

130. Holden BA, Williams L, Zantos SG. The etiology of transient endothelial changes in the human cornea. *Investig. Ophthalmol. Vis. Sci.* 1985;26(10):1354-1359.

131. Giasson C, Bonanno JA. Corneal epithelial and aqueous humor acidification during in vivo contact lens wear in rabbits. *Investig. Ophthalmol. Vis. Sci.* 1994;35(3):851-861.

132. Setälä K, Vasara K, Vesti E, Ruusuvaara P. Effects of long-term contact lens wear on the corneal endothelium. *Acta Ophthalmol. Scand.* 1998;76(3):299-303.

133. Lu F, Xu S, Qu J, *et al*. Central corneal thickness and corneal hysteresis during corneal swelling induced by contact lens wear with eye closure. *Am. J. Ophthalmol.* 2007;143(4):616-22.

134. Franco S, Lira M. Biomechanical properties of the cornea measured by the Ocular Response Analyzer and their association with intraocular pressure and the central corneal curvature. *Clin. Exp. Optom.* 2009;92(6):469-475.

135. Chen D, Lam AKC, Cho P. A pilot study on the corneal biomechanical changes in short-term orthokeratology. *Ophthalmic Physiol. Opt.* 2009;29(4):464-471.

136. Nieto-Bona A, González-Mesa A, Villa-Collar C, Lorente-Velázquez A. Biomechanical properties in corneal refractive therapy during adaptation period and after treatment interruption: A pilot study. *J. Optom.* 2012;5(4):164-170.

137. Golebiowski B, Papas EB, Stapleton F. Corneal and conjunctival sensory function: The impact on ocular surface sensitivity of change from low to high oxygen transmissibility contact lenses. *Investig. Ophthalmol. Vis. Sci.* 2012;53(3):1177-1181.

138. Polse KA. Etiology of corneal sensitivity changes accompanying contact lens wear. *Invest Ophthalmol Vis Sci* 1978;17(12):1202.

139. Situ P, Simpson TL, Jones LW, Fonn D. Effects of silicone hydrogel contact lens wear on ocular surface sensitivity to tactile, pneumatic mechanical, and chemical stimulation. *Investig. Ophthalmol. Vis. Sci.* 2010;51(12):6111-6117.

140. Chalmers RL, Dillehay S, Long B, *et al*. Impact of previous extended and daily wear schedules on signs and symptoms with high Dk lotrafilcon A lenses. *Optom. Vis. Sci.* 2005;82(6):549-554.

141. Papas EB, Vajdic CM, Austen R, Holden BA. High-oxygen-transmissibility soft contact lenses do not induce limbal hyperaemia. *Curr. Eye Res.* 1997;16(9):942-948.

142. Papas EB. The role of hypoxia in the limbal vascular response to soft contact lens wear. *Eye Contact Lens* 2003;29(1 Suppl):S72-S74; discussion S83-S84, S192-S194.

143. Maldonado-Codina C, Morgan PB, Schnider CM, Efron N. Short-term physiologic response in neophyte subjects fitted with hydrogel and silicone hydrogel contact lenses. *Optom. Vis. Sci.* 2004;81(12):911-921.

144. Glasson MJ, Stapleton F, Keay L, Willcox MDP. The effect of short term contact lens wear on the tear film and ocular surface characteristics of tolerant and intolerant wearers. *Contact Lens Anterior Eye* 2006;29(1):41-47.

145. De la Jara PL, Papas E, Diec J, Naduvilath T, Willcox MDP, Holden BA. Effect of Lens Care Systems on the Clinical Performance of a Contact Lens. *Optom. Vis. Sci.* 2013;90(4):344-350.

146. Duench S, Simpson T, Jones LW, Flanagan JG, Fonn D. Assessment of variation in bulbar conjunctival redness, temperature, and blood flow. *Optom. Vis. Sci.* 2007;84(6):511-516.

147. Junghans BM, Collin HB. The limbal vascular response to corneal injury. An autoradiographic study. *Cornea* 1989;8(2):141-149.

148. Miyashita H, Higa K, Kato N, *et al*. Hypoxia enhances the expansion of human limbal epithelial progenitor cells in vitro. *Investig. Ophthalmol. Vis. Sci.* 2007;48(8):3586-3593.

149. Woods J. Conjunctival Responses to Contact Lens Wear. *Contact Lens Spectr.* 2015;30:34-36,38.

150. Papas E. On the relationship between soft contact lens oxygen transmissibility and induced limbal hyperaemia. *Exp. Eye Res.* 1998;67(2):125-131.

151. Michaud L, Giasson CJ. Overwear of Contact Lenses : Increased Severity. *Optom. Vis. Sci.* 2002;79(3):184-192.

152. Holden BA, La Hood D, Grant T, *et al*. Gram-negative bacteria can induce contact lens related acute red eye (CLARE) responses. *CLAO J* 1996;22(1):47-52.

153. Papas EB. Lissamine Green Versus Rose Bengal. *Contact Lens Spectr.* 2014;29:16-17.

154. Morgan PB, Maldonado-Codina C. Corneal staining: Do we really understand what we are seeing? *Contact Lens Anterior Eye* 2009;32(2):48-54.

155. Maïssa C, Guillon M, Garofalo RJ. Contact Lens–Induced Circumlimbal Staining in Silicone Hydrogel Contact Lenses Worn on a Daily Wear Basis. *Eye Contact Lens* 2012;38(1):16-26.

156. Guillon M, Maissa C. Bulbar conjunctival staining in contact lens wearers and non lens wearers and its association with symptomatology. *Contact Lens Anterior Eye* 2005;28(2):67-73.

157. Keir N, Woods J, Sickenberger W. The Conjunctival Response to Soft Contact Lens Wear: A Practical Guide. Optometry in Practice (UK).; 2010.

158. Markoulli M, Francis IC, Yong J, *et al*. A histopathological study of bulbar conjunctival flaps occurring in 2 contact lens wearers. *Cornea* 2011;30(9):1037-1041.

159. Lofstrom T, Kruse A. A conjunctival response to silicone hydrogel lens wear. *Contact Lens Spectr.* 2005;20:42-44.

160. Graham AD, Truong TN, Lin MC. Conjunctival epithelial flap in continuous contact lens wear. *Optom. Vis. Sci.* 2009;86(4):e324-e331.

161. Santodomingo-Rubido J, Wolffsohn J, Gilmartin B. Conjunctival Epithelial Flaps With 18 Months of Silicone Hydrogel Contact Lens Wear. *Eye Contact Lens* 2008;34(1):35-38.

162. Lin MC, Yeh TN. Mechanical complications induced by silicone hydrogel contact lenses. *Eye Contact Lens* 2013;39(1):115-24.

163. Nemeth J, Fodor E, Lang Z, *et al*. Lid-parallel conjunctival folds (LIPCOF) and dry eye: a multicentre study. *Br J Ophthalmol* 2012;96(11):1380-1385.

164. Hall PA, Watt FM. Stem cells: the generation and maintenance of cellular diversity. *Development* 1989;106(4):619-633.

165. Jeng BH, Halfpenny CP, Meisler DM, Stock EL. Management of focal limbal stem cell deficiency associated with soft contact lens wear. *Cornea* 2011;30(1):18-23.

166. Chan CC, Holland EJ. Severe Limbal Stem Cell Deficiency from Contact Lens Wear: Patient Clinical Features. *Am. J. Ophthalmol.* 2012;155(3):544-549.e2.

167. Doughty MJ. Contact lens wear and the goblet cells of the human conjunctiva — A review. *Contact Lens Anterior Eye* 2011;34(4):157-163..

168. Greiner J V, Covington HI, Korb DR, Allansmith MR. Conjunctiva in asymptomatic contact lens wearers. *Am. J. Ophthalmol.* 1978;86:403-413.

169. Greiner J V, Allansmith MR. Effect of contact lens wear on the conjunctival mucous system. *Ophthalmology* 1981;88:821-832.

170. Yasueda S, Yamakawa K, Nakanishi Y, Kinoshita M, Kakehi K. Decreased mucin concentrations in tear fluids of contact lens wearers. *J. Pharm. Biomed. Anal.* 2005;39(1-2):187-95.

171. Pisella PJ, Malet F, Lejeune S, *et al*. Ocular surface changes induced by contact lens wear. *Cornea* 2001;20(8):820-5.

172. Hirji N, Scott J, Sabel A. Conjunctival cytology in hard and soft contact lens wear. *Ophthalmic Physiol. Opt.* 1985;5(3):333-335.

173. Simon P, Jaison SG, Chopra SK, Jacob S. Conjunctivl Impression Cytology in Contact Lens Wearers. *Indian J Ophthalmol* 2002;50:301-306.

174. Cakmak SS, Unlü MK, Karaca C, Nergiz Y, Ipek S. Effects of soft contact lenses on conjunctival surface. *Eye Contact Lens* 2003;29(4):230-3.

175. Lievens CW, Connor CG, Murphy H. Comparing goblet cell densities in patients wearing disposable hydrogel contact lenses versus silicone hydrogel contact lenses in an extended-wear modality. *Eye Contact Lens* 2003;29(4):241-4.

176. Connor CG, Campbell JB, Steel SA, Burke JH. The effects of daily wear contact lenses on goblet cell density. *J. Am. Optom. Assoc.* 1994;65(11):792-4.

177. Adar S, Kanpolat A, Sürücü S UO. Conjunctival impression cytology in patients wearing contact lenses. *Cornea* 1997;16(3):289-94.

178. Doughty MJ, Naase T. Nucleus and cell size changes in human bulbar conjunctival cells after soft contact lens wear , as assessed by impression cytology. *Contact Lens Anterior Eye* 2008;31:131-140.

179. Knop E, Brewitt H. Induction of conjunctival epithelial alterations by contact lens wearing. *Ger J Ophthalmol* 1992;1(3-4):125-34.

180. Tomatir DK, Erda N, Gürlü VP. Effects of different contact lens materials and contact lens-wearing periods on conjunctival cytology in asymptomatic contact lens wearers. *Eye Contact Lens* 2008;34(3):166-8.

181. Dumbleton KA, Chalmers RL, Richter DB, Fonn D. Changes in myopic refractive error with nine months' extended wear of hydrogel lenses with high and low oxygen permeability. *Optom. Vis. Sci.* 1999;76(12):845-849.

182. Jalbert I, Stretton S, Naduvilath T, Holden B, Keay L, Sweeney D. Changes in Myopia with Low-Dk Hydrogel and High-Dk Silicone Hydrogel Extended Wear. *Optom. Vis. Sci.* 2004;81(8):591-596.

183. Dumbleton K, Keir N, Moezzi A, Feng Y, Jones L, Fonn D. Objective and subjective responses in patients refitted to daily-wear silicone hydrogel contact lenses. *Optom. Vis. Sci.* 2006;83(10):758-768.

184. Gogniat F, Steinegger D, Nosch D, Joos R, Goldschmidt M. The Accuracy of Dynamic Contour Tonometry. *Optom. Vis. Sci.* 2013;90(2):125-130.

185. McMonnies CW. Noncontact tonometry through soft contact lenses. *Am. J. Optom. Physiol. Opt.* 1986;63(12):948-951.

186. Schollmayer P, Hawlina M. Effect of Soft Contact Lenses on the Measurements of Intraocular Pressure with Non-Contact Pneumotonometry. *Klin. Monbl. Augenheilkd.* 2003;220(12):840-842.

187. Zeri F, Lupelli L, Formichella P, Masci C, Fletcher R. Goldmann applanation tonometry over daily disposable contact lens: Accuracy and safety of procedure. *Contact Lens Anterior Eye* 2007;30:233–238.

188. Patel S, Stevenson G. Influence of lens material and intra-ocular pressure on the outcome of non-contact tonometry over soft contact lenses. *Contact Lens Anterior Eye* 2009;32(2):68-72.

189. Liu Y-C, Huang J-Y, Wang I-J, Hu F-R, Hou Y-C. Intraocular pressure measurement with the noncontact tonometer through soft contact lenses. *J. Glaucoma* 2011;20(3):179-182.

190. Zeri F, Calcatelli P, Donini B, Lupelli L, Zarrilli L, Swann PG. The effect of hydrogel and silicone hydrogel contact lenses on the measurement of intraocular pressure with rebound tonometry. *Contact Lens Anterior Eye* 2011;34(6):260-265.

191. Fırat PG, Cankaya C, Doganay S, *et al*. The influence of soft contact lenses on the intraocular pressure measurement. *Eye* 2012;26(2):278-282.

192. Allen RJ, Dev Borman A, Saleh GM. Applanation tonometry in silicone hydrogel contact lens wearers. *Contact Lens Anterior Eye* 2007;30(5):267-269.

193. Schornack M, Rice M, Hodge D. Tonopen XL Assessment of Intraocular Pressure Through Silicone Hydrogel Contact Lenses. *Eye Contact Lens Sci. Clin. Pract.* 2012;38(5):270-273.

194. Boyraz S, Güngör I. The effects of the modulus of the lens material on intraocular pressure measurement through soft contact lenses. *Ir. J. Med. Sci.* 2013;182(3):331-335.

195. Anton A, Neuburger M, Böhringer D, Jordan JF. Comparative measurement of intraocular pressure by Icare tonometry and Airpuff tonometry in healthy subjects and patients wearing therapeutic soft contact lenses. *Graefe's Arch. Clin. Exp. Ophthalmol.* 2013;251(7):1791-1795.

196. Harada Y, Hirose N, Kubota T, Tawara A. The influence of central corneal thickness and corneal curvature radius on the intraocular pressure as measured by different tonometers: noncontact and goldmann applanation tonometers. *J. Glaucoma* 2008;17(8):619-25.

197. Saleh TA, Adams M, McDermott B, Claridge KG, Ewings P. Effects of central corneal thickness and corneal curvature on the intraocular pressure measurement by Goldmann applanation tonometer and ocular blood flow pneumatonometer. *Clin. Experiment. Ophthalmol.* 2006;34(6):516-20.

198. Mahjoob M, Azimi A, Momeni-moghadam H, Mohamadian M, Yousofi R. The Effect of Various Contact Lenses on Intraocular Pressure Measurement by Goldman Tonometer. *Zahedan J Res Med Sci* 2014;16(6):33-35.

199. Oh JH, Yoo C, Kim YY, Kim HM SJ. The effect of contact lens-induced corneal edema on Goldmann applanation tonometry and dynamic contour tonometry. *Graefes Arch Clin Exp Ophthalmol* 2009;247(3):371-5.

200. Briggs ST. Contrast sensitivity assessment of soft contact lens wearers. *Int. Contact Lens Clin.* 1998;25(98):99-102.

201. De Fez MD, Luque MJ, Viqueira V. Enhancement of contrast sensitivity and losses of chromatic discrimination with tinted lenses. *Optom. Vis. Sci.* 2002;79(9):590-597.

202. Ortiz C, Jimenez R. Optical Quality and Vision with Iris-Coloring Soft Contact Lenses. *Optom. Vis. Sci.* 2014;91(5):564-569.

203. Grey CP. Changes in contrast sensitivity during the first hour of soft lens wear. *Am. J. Optom. Physiol. Opt.* 1986;63:702-707.

204. Grey CP. Changes in contrast sensitivity during the first six months of soft lens wear. *Am. J. Optom. Physiol. Opt.* 1987;64:768-774.

205. Soni PS, Patel R, Carlson RS. Is binocular contrast sensitivity at distance compromised with multifocal soft contact lenses used to correct presbyopia? *Optom. Vis. Sci.* 2003;80(7):505-514.

206. Porisch E. Football players' contrast sensitivity comparison when wearing amber sport-tinted or clear contact lenses. *Optometry* 2007;78:232-235.

207. Carracedo G, Carballo J, Loma E, Felipe G, Cacho I. Contrast sensitivity evaluation with filter contact lenses in patients with retinitis pigmentosa: A pilot study. *J. Optom.* 2011;4(4):134-139.

208. Dalcoll MW, Alves MR, Barreto J, Yamane IDS, Bechara S, Mukai A. Evaluation of optical performance of soft contact lenses in myopic correction. *Arq. Bras. Oftalmol.* 2008;71(6 suppl):37-41.

209. Barth B, Alves MR, Kara-José N. Visual performance in myopic correction with spectacles and soft contact lenses. *Arq. Bras. Oftalmol.* 2008;71(1):90-96.

210. Belda-Salmerón L, Ferrer-Blasco T, Albarrán-Diego C, Madrid-Costa D, Montés-Micó R. Diurnal variations in visual performance for disposable contact lenses. *Optom. Vis. Sci.* 2013;90(7):682-90.

CHAPTER 3. **A**

GOBLET CELL DENSITY ASSOCIATION WITH TEAR FUNCTION AND OCULAR SURFACE PHYSIOLOGY

Highlights

- **Goblet cell density (GCD) was determined on the bulbar conjunctiva of asymptomatic subjects.**
- **Correlation of GCD with tear function and ocular surface physiology was evaluated.**
- **GCD was found significantly correlated with tear break-up time.**
- **No correlation of GCD with Schirmer scores, conjunctival redness and corneal staining was found.**

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3A.1 Introduction

Normal tear film maintains an optically uniform ocular surface, protects the eyes from infections and environmental hazards, washes out cellular debris and foreign bodies and sustains ocular comfort and a healthy corneal epithelium. Mucin, the innermost layer of the tear, changes the hydrophobic corneal surface into hydrophilic. It also makes the corneal surface smooth and helps to balance tear film on the anterior ocular surface. Conjunctival goblet cells produce mucin-MUC5AC and it is supposed to be one of the main gel forming substances in the tears.[1] Goblet cells are balloon shaped cells with eccentric nucleus found in the superficial layers of the conjunctiva.

Assessment of goblet cell on the ocular surface is important as it is a helpful diagnostic tool for many eye conditions. Goblet cell density (GCD) was found lower in eyes with ocular surface disease in comparison to normal eyes.[2] The number of goblet cells decreases with contact lens wear.[3] Torricelli and co-workers found an increase in GCD due to air pollution.[4] Doughty reviewed 24 different studies which reported the effects of contact lens wear on the goblet cells of the human conjunctiva and found that there has been limited consistency in the technique or the method of reporting the results across the various studies justifying the differences obtained.[3] Additionally, GCD in conjunctival surface varies highly from one region to another and from one person to another with a range from 24 to 2226 cells/mm^2 for average values.[2] Torricelli *et al*. found higher GCD in tarsal conjunctiva in comparison to bulbar conjunctiva.[4] Connor *et al*. reported that GCD is higher in male than in females and in female, it is higher in oral contraceptive users.[5] However, Tomlinson *et al*. found no relationship between tear physiology and oral contraceptives.[6] Yeo *et al*. conducted impression cytology in 40 healthy Chinese subjects.[7] They did not find any correlation between GCD and Schirmer's score, non-invasive break-up time (NIBUT) and tear break-up time (TBUT).

Conjunctival impression cytology (CIC) is a simple and less invasive method (over a conventional surgical biopsy) to assess the conjunctival health including conjunctival epithelial metaplasia and goblet cell density.[8] Goblet cells are easily visualized through light microscope by staining their mucus content with Periodic Acid-Schiff (PAS).[9] During the last decade, CIC has been applied enormously as a useful diagnostic aid for a wide variety of ocular surface pathologies

and has greatly contributed to the understanding of ocular surface conditions.[8] It implies the collection of cells from the conjunctival surface via a special type of filter paper (or sometimes disc) by impression on the surface and examination of the superficial layer(s) with different types of staining. It can be done with or without anesthesia.[9]

Two different approaches have been used to evaluate the goblet cells obtained in a CIC sample: a direct assessment of the number of goblet cells or an indirect evaluation (based on the assignment of a grade).[3] Estimation of GCD has been carried out by different ways. Although 'number of goblet cells per millimeter', 'number of goblet cells per high power field' and 'number of goblet cells per 100 basal cells' have been reported in different studies; 'number of goblet cells/mm^2 is the most used unit.[2] Generally, the numbers of cells are counted in a unit field of view of the microscope and cells/mm^2 is calculated.

Limited numbers of studies have been conducted to determine the relationship between conjunctival GCD with ocular surface physiology and tear function tests. The aims of this study were to investigate GCD in eyes of asymptomatic subjects and to determine its correlation with limbal/bulbar redness, corneal staining, Schirmer's score, NIBUT and TBUT.

3A.2 Methods

This was a cross-sectional study conducted in University of Minho, Portugal. Each subject signed a consent form after the procedures, time duration, possible consequences and other details of the study were explained. This study was ethically approved by Ethical Committee of School of Science, University of Minho and tenets of declaration of Helsinki were followed.

Thirty-five volunteers with age 18-35 years and minimum best corrected visual acuity of 6/6 in each eye, were included in the study. The sample size was calculated to warrant the 70% statistical power with 0.05 significance level which was estimated on the basis of our preliminary data. Subjects with past history of contact lens wear were excluded from the study. Each subject filled up McMonnies questionnaire and those with score more than 14 were excluded from the study.[10,11]

Schirmer's test was done without anesthesia with a commercially available paper strip (Sno strips, *Laboratoire* Chauvin, France). The strip was inserted into the lower temporal conjunctival sac and the wet length of the paper was measured on millimeter after five minutes. Eyes were closed during the test to get more reliable data.[12] NIBUT was performed with Tearscope Plus (Keeler Instruments Ltd, Windsor). By applying fluorescein dye, TBUT was measured as the time interval between the subject blink and the first appearance of a black spot or line on the cornea observed by slit lamp with cobalt blue light and a Wratton-12 yellow filter. Schirmer's score less than 10mm[13,14] and TBUT score less than 10sec,[14,15] were considered as low values.

Bulbar conjunctiva and limbus were divided into four regions: nasal, temporal, superior and inferior and the cornea was divided into five regions: central, nasal, temporal, superior and inferior as described in the CLEK study.[16] Bulbar and limbal redness and corneal staining were graded applying Efron grading system with score 0-4.[17] Each of the grading's was done on 0.1 step to optimize grading sensitivity.[18] The average score was used for data analysis.

Impression cytology was performed on the superior bulbar conjunctiva on both eyes of each selected subject.[9] Nitrocellulose Millipore[8] MF™-Membrane filter (MILLIPORE, Ireland) with pore size 0.45µm was used without application of topical anesthesia.[19,20] Briefly, a circular paper of diameter 13 mm was cut into two equal semi-circular pieces. The semi-circular piece of a filter was placed on the superior bulbar conjunctiva, 1-2 mm away from the limbus and removed in peeling motion. The paper was dried and fixed with 96% ethanol for 15 minutes in a 24 well plate. The paper was then stained with PAS, Haemotoxylin and Eosin and dried in ascending concentration of ethanol.[21] The slides were prepared by dissolving the paper with Xylene and mounted with Xylol. The slides were observed by light microscopy with total magnification 100X and 400X. Goblet cells were counted in the higher magnification with an area of 0.16 mm^2 (total magnification of 400X) and GCD was calculated as the number of cells per square millimeter. This procedure was repeated in three random fields of areas and the average was used in the analysis.

Data were analyzed with Statistical Software (SPSS 22, IBM Corp., Armonk, NY). Kolmogorov-Smirnov test was done to find out the normality of the variables. Parametric tests were applied for normally distributed variables and non-parametric tests were applied for others.

Correlation between two variables was found out by Spearman test, and the association with gender or oral medication was found out by one-way ANOVA with Post-hoc. A p value of less than 0.05 was considered as statistically significant. Data of the right eyes were used for the analysis.

3A.3 Results

Among the 35 asymptomatic subjects enrolled in this study, 23 (65.7%) were females. The mean age of the subjects was 23.8±3.6 years. The majority (91.4%) of the subjects were Caucasians. None of the subjects had worn contact lens before the study.

After conjunctival impression cytology, the cells imprinted in the paper filter were stained and visualized by optical microscopy. The goblet cells were easily identified under the microscope due to their dark pink colour after PAS staining [Figure 3A.1].

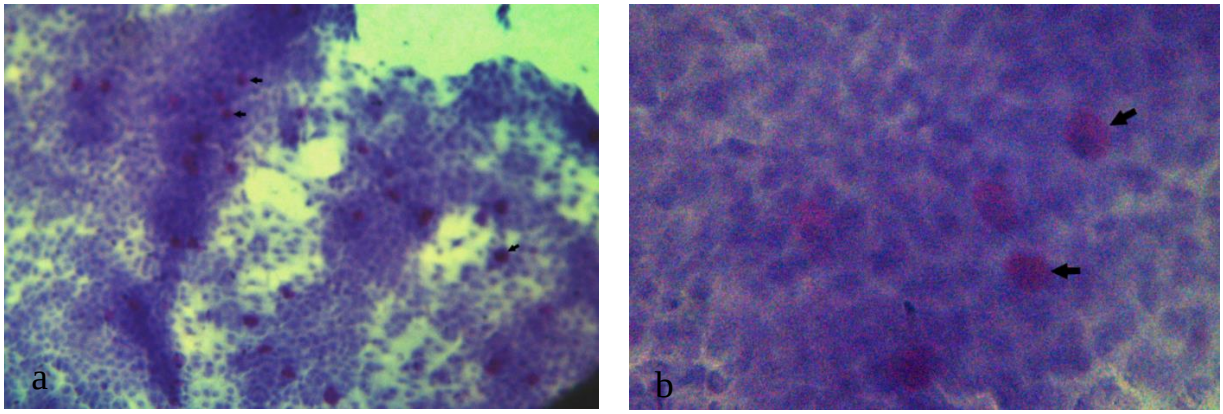


Figure 3A.1. Conjunctival impression cytology of one individual, showing dark pink stained goblet cells (arrow) and light stained epithelial cells a. 100X, b. 400X total magnification.

The conjunctival cytology was observed with 100X and 400X total magnification with light microscope [Figure 3A.1] and the goblet cells were counted. Descriptive statistics of the variables are shown in Table 3A.1. A high variation in GCD was observed with a mean value of 151 cells/mm^2 and standard deviation of 122 cells/mm^2.

Table 3A.1. Descriptive statistics of the variables

	Mean	SD	Range
GCD (cells/mm^2)	151	122	18-522
Schirmer score (mm)	22.7	11.6	3-45
NIBUT (s)	20.0	14.4	7-60
TBUT (s)	12.4	7.2	5-36
Bulbar redness	0.3	0.23	0-0.9
Limbal redness	0.4	0.26	0-1.0
Corneal staining	0.2	0.13	0-0.5

GCD – goblet cell density, NIBUT – non-invasive tear break up time, TBUT – tear break up time, SD – standard deviation

The correlation between each variable is shown in Table 3A.2. As it can be seen, no correlation between GCD and Schirmer score ($r = 0.05$, $p = 0.75$) was found. A positive correlation with the NIBUT was observed, but not statistically significant ($r = 0.224$, $p = 0.195$). The GCD was found to be positively correlated with TBUT ($r = 0.338$, $p = 0.042$).

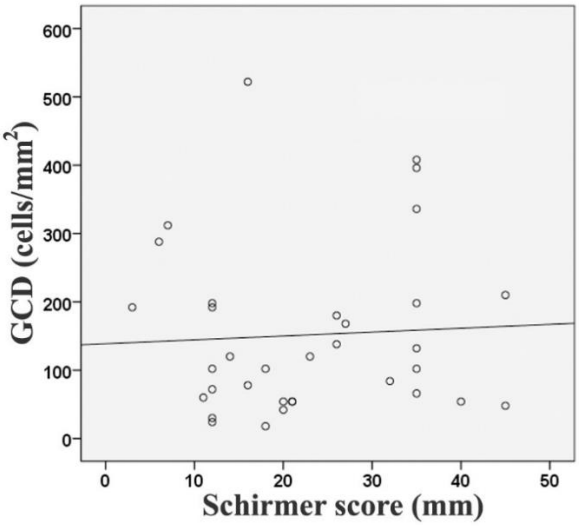
Figure 3A.2 shows the scatter plot of GCD versus Schirmer score and TBUT. The inverse correlation of GCD with conjunctival bulbar as well as with limbal redness and corneal staining could not reach a statistically significant level ($p > 0.05$). Corneal staining was found inversely correlated with Schirmer score and positively correlated with bulbar/limbal redness ($p < 0.05$).

Table 3A.2. Correlation between variables (Spearman's rho values)

	GCD (cells/mm^2)	**Schirmer score (mm)**	**NIBUT (s)**	**TBUT (s)**	**Bulbar redness**	**Limbal redness**	**Corneal staining**
GCD (cells/mm^2)	1						
Schirmer score (mm)	0.05 (p=0.750)	1					
NIBUT (s)	0.224 (p=0.195)	0.2221 (p=0.209)	1				
TBUT (s)	0.338 (p=0.042)	0.038 (p=0.832)	0.731 (p=0.000)	1			
Bulbar redness	-0.264 (p=0.126)	-0.074 (p=0.685)	0.144 (p=0.410)	0.242 (p=0.154)	1		
Limbal redness	-0.328 (p=0.054)	-0.0063 (p=0.954)	0.154 (p=0.377)	0.196 (p=0.259)	0. 901 (p=0.000)	1	
Corneal staining	-0.301 (p=0.079)	-0.382 (p=0.026)	0.010 (p=0.953)	-0.083 (p=0.636)	0.405 (p=0.008)	0.456 (p=0.006)	1

GCD – goblet cell density, NIBUT – non-invasive tear break-up time, TBUT – tear break up time

a.



b.

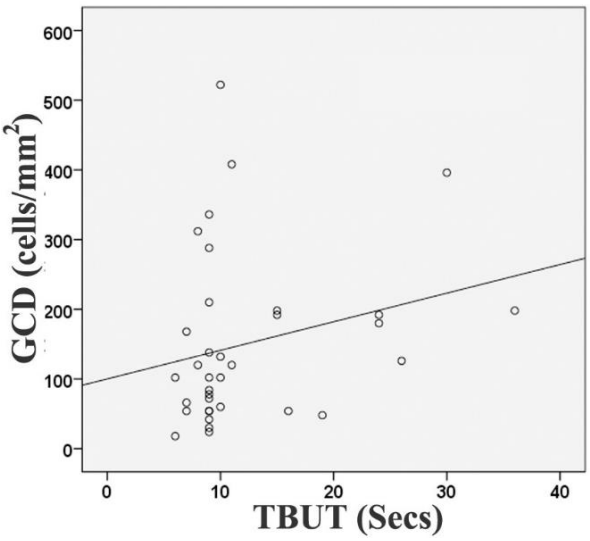


Figure.3A.2. Plotting of GCD with a). Schirmer score and b). TBUT
[GCD: goblet cell density; TBUT: tear break-up time].

As shown in Table 3A.3, eyes with low TBUT had statistically significant lower GCD ($p = 0.043$), however, the difference in GCD, in eyes with low and high Schirmer values, could not reach to statistically significant level ($p = 0.058$).

Although GCD decreased with age, this inverse relationship was not statistically significant ($p = 0.162$). There was no difference in GCD between oral contraceptive user women or other women and men ($p > 0.05$).

Table 3A.3. Goblet cell density with different tear values

		GCD	P values
TBUT	<10Sec	118	0.043
	≥10 Sec	195	
Schirmer score	<10 mm	264	0.058
	≥10 mm	141	

3A.4 Discussion

Goblet cells produce mucin which is one of the most important components of the tears. Number of the goblet cells may affect vision, ocular comfort and ocular health. This study determined the GCD on the superior palpebral conjunctiva by CIC in asymptomatic subjects and its relation with other tear functions as well as ocular surface physiology.

The average GCD found was 151±122 cells/mm^2 (Table 1). The large standard deviation indicates that there was a huge variation in GCD, as previously reported.[2] Contrary to what was found in the present study, Zuazo *et al*.[22] found GCD of 497 cells/mm^2 in normal subjects and 310 cells/mm^2 in subjects with dry eyes, and Torricelli *et al*.[4] found GCD of 521 cells/mm^2 in bulbar conjunctiva of normal subjects. Similarly, in a recent study conducted in India, Kumar *et al*. found GCD of 1462 cells/mm^2 in normal eyes.[23] The lower GCD obtained in this study may be due to the fact that no pressure was applied during impression cytology process, unlike those authors.[4,23] In that way, only superficial layer(s) were removed and goblet cells in the deep layer may not be collected in the sample. Moreover, subjects had lower values of tear function tests in comparison to other study.[24] It was because subjects with a low level of tear function test were also included since they were apparently normal and without any ocular symptoms. Lower GCD may be due to mild or moderate dry eyes in these subjects as it was shown by Murube and Rivas.[25] It was also found lower GCD in eyes with unstable tears (TBUT < 10 secs) but significant differences in eyes with low aqueous production (Schirmer < 10mm) could not be found (Table 3A.3), which may be due to the fact that bulbar conjunctiva which is covered by lids, is less affected by aqueous deficient

eye.[26] However, some authors found even low number of goblet cells in normal subjects.[27] Doughty in a meta-analysis of CIC in normal eyes concluded that GCD is highly varied with range 24 cells/mm^2 to 2226 cells/mm^2.[2]

It's not surprising that no correlation was found with GCD and Schirmer's score. Consistent to this study, Yeo *et al.*[7] and Paschides *et al.*[28] also did not find any relation of GCD with phenol red thread test score. It might be because there is little or no direct role of goblet cells on the aqueous portion of the tears.

There was a positive correlation with GCD and NIBUT although it was not significant ($p = 0.195$). Nevertheless, there was a significant positive correlation between GCD and TBUT. This shows that the number of goblet cells can affect the TBUT since mucin produced in goblet cells is important for the tear stability. However, the correlation was weak between GCD and TBUT. Yeo *et al.*[7] and Paschides *et al.*[28] did not find any correlation of GCD with NIBUT and TBUT. Yeo *et al*. suggested that TBUT depends not only upon MUC5AC which is produced by goblet cells but also with other mucin produced by other conjunctival cells.[7]

There was no correlation of GCD with conjunctival redness and corneal staining. However, the inverse correlation of GCD with limbal redness and corneal staining was close to statistically significant level. It shows the importance of the goblet cells on maintenance of ocular health. It may be due to the fact that insufficient mucin level in tears makes the ocular surface dry and vulnerable to inflammation/infection.

Although age and gender might be considered as factors that could affect GCD, in this study, there was no significant relation of age and gender with GCD. Doughty reviewed the studies published regarding goblet cells of the normal human bulbar conjunctiva and their assessment by CIC sampling but could not find any predictable effect of age on GCD.[2] Aqueous deficient dry eye was found correlated to age and gender,[29] but GCD did not. This highlights that there is no role of goblet cells on aqueous deficient dry eyes. Moreover, there was no difference in GCD between oral contraceptive user women and others. In contrary to this finding, Connor *et al*. showed higher GCD in oral contraceptives users.[5]

There were some limitations in this study. Goblet cells were counted in the higher power field of view (with 400X magnification), and the chance of error is higher with higher power field.[30] However, to minimize this effect, goblet cells were computed in three random areas and average was used for analysis.

Use of anaesthesia is not allowed for optometrists in many European countries including Portugal. Anaesthesia was not used during the impression cytology; pressure was not applied on the filter paper; so it is assumed that only superficial layer(s) was/were attached on the filter paper. Special care was taken to count only goblet cells but not the goblet cell secretions.[9] Egbert *et al.* observed that the density of goblet cells were more likely to be observed across regions of the filter where more than one layer of cells had been removed.[9] This sampling variability may reflect differences in manual pressure exerted onto the bulbar conjunctiva after application of a small piece of filter paper. So, differences in GCD could occur because of sample size variation and this could include both the most superficial cells as well slightly deeper and even cells close to the basal layer.

In conclusion, GCD in superior bulbar conjunctiva varied highly in asymptomatic subjects. There was no correlation of goblet cells with aqueous portion of tears however, TBUT was found correlated with GCD. GCD did not correlate with age and gender of the subjects.

References

1. Gipson IK, Argüeso P. Role of mucins in the function of the corneal and conjunctival epithelia. *Int. Rev. Cytol.* 2003;231:1-49.

2. Doughty MJ. Goblet Cells of the Normal Human Bulbar Conjunctiva and Their Assessment by Impression Cytology Sampling. *Ocul. Surf.* 2012;10(3):149-169.

3. Doughty MJ. Contact lens wear and the goblet cells of the human conjunctiva — A review. *Contact Lens Anterior Eye* 2011;34(4):157-163.

4. Torricelli AA, Miranda, Matsuda M, *et al*. Effects of ambient levels of traffic-derived air pollution on the ocular surface : Analysis of symptoms, conjunctival goblet cell count and mucin 5AC gene expression. *Environ. Res.* 2014;131(2):59-63.

5. Connor C, Flockencier L, Hall C. The influence of gender on the ocular surface. *J Am Optom Assoc* 1999;70(3):182-6.

6. Tomlinson A, Pearce EI, Simmons PA, Blades K. Effect of oral contraceptives on tear physiology. 2001;21(1).

7. Yeo ACH, Carkeet A, Carney LG, Keng M, Yap H. Relationship between goblet cell density and tear function tests. *Ophthal Physiol Opt* 2003;23:87-94.

8. Calonge M, Enrı A, Salamanca D, *et al*. Impression cytology of the ocular surface : a review. *Exp. Eye Res.* 2004;78:457-472.

9. Egbert PR, Lauber S MD. A simple conjunctival biopsy. *Am. J. Ophthalmol.* 1977;84(6):798-801.

10. McMonnies CW. Key questions in a dry eye history. *J. Am. Optom. Assoc.* 1986;57:512-517.

11. Moore JE, Graham JE, Goodall E a, *et al*. Concordance between common dry eye diagnostic tests. *Br. J. Ophthalmol.* 2009;93:66-72.

12. Serin D, Karsloğlu S, Kyan A, Alagöz G. A simple approach to the repeatability of the Schirmer test without anesthesia: eyes open or closed? *Cornea* 2007;26(8):903-6.

13. Jordan A, Baum J. Basic Tear Flow. *Ophthalmology* 1980;87(9):920-930.

14. Vitali C, Moutsopoulos HM, Bombardieri S. The European Community Study Group on diagnostic criteria for Sjögren's syndrome. Sensitivity and specificity of tests for ocular and oral involvement in Sjögren's syndrome. *Ann. Rheum. Dis.* 1994;53(10):637-47.

15. Zeev MS-B, Miller DD, Latkany R. Diagnosis of dry eye disease and emerging technologies. *Clin. Ophthalmol.* 2014;8:581-90.

16. Barr JT, Schechtman KB, Fink B a, *et al*. Corneal scarring in the Collaborative Longitudinal Evaluation of Keratoconus (CLEK) Study: baseline prevalence and repeatability of detection. *Cornea* 1999;18(1):34-46.

17. Nathon E. Efron Grading Scales for Contact Lens Complications. In: Efron N, Ed. Contact Lens Complications. Oxford: Butterworth- Heinnemann; 2004:239-43.

18. Bailey IL, Bullimore M, Raasch TW, Taylor HR. Clinical grading and the effects of scaling. *Invest. Ophthalmol. Vis. Sci.* 1991;32(2):422-32.

19. Blades K, Doughty MJ, Patel S. Pilot Study on the Use of Impression Cytology Specimens for Quantitative Assessment of the Surface Area of Bulbar Conjunctival Cells. *Optom. Vis. Sci.* 1998;75(8):591-9.

20. Thatcher RW, Darougar S JB. Conjunctival impression cytology. *Arch Ophthalmol* 1977;95(4):678-81.

21. Rivas L, Oroza MA, Perez-Esteban A M-CJ. Topographical distribution of ocular surface cells by the use of impression cytology. *Acta Ophthalmol* 1991;69(3):371-6.

22. Zuazo F, López-Ponce D, Salinas-Toro D, *et al*. Conjunctival impression cytology in patients with normal and impaired OSDI scores. *Arch. Soc. Esp. Oftalmol.* 2014;89(10):391-6.

23. Kumar P, Bhargava R, Kumar M, Ranjan S, Kumar M, Verma P. The Correlation of Routine Tear Function Tests and Conjunctival Impression Cytology in Dry Eye Syndrome. *Korean J Ophthalmol* 2014;28(2):122-129.

24. Dogru M, Katakami C, Nakagawa N, Tetsumoto K, Yamamoto M. Impression Cytology in Atopic Dermatitis. 1998:1478-1484.

25. Murube J, Rivas L. Impression cytology on conjunctiva and cornea in dry eye patients establishes a correlation between squamous metaplasia and dry eye clinical severity. *Eur. J. Ophthalmol.* 2003;13(2):115-27.

26. Tseng S. Staging of conjunctival squamous metaplasia by impression cytology. *Ophthalmology* 1985;92(6):728-33.

27. Adar S, Kanpolat A, Sürücü S UO. Conjunctival impression cytology in patients wearing contact lenses. *Cornea* 1997;16(3):289-94.

28. Paschides CA, Petroutsos G, Psilas K. Correlation of conjunctival impression cytology results with lacrimal function and age. *Acta Ophthalmol.* 1991;69(4):422-5.

29. Albietz JM. Prevalence of dry eye subtypes in clinical optometry practice. *Optom. Vis. Sci.* 2000;77(7):357-63.

30. Doughty MJ. Sampling area selection for the assessment of goblet cell density from conjunctival impression cytology specimens. *Eye Contact Lens* 2012;38(2):122-9.

CHAPTER 3 B

EFFECTS OF THREE MONTHS OF SOFT CONTACT LENS WEAR ON CONJUNCTIVAL CYTOLOGY

Highlights

- **Change in goblet cell density (GCD) and epithelial cell grading was investigated on the superior bulbar conjunctiva of neophyte soft contact lens (SCL) wearers.**
- **GCD reduced significantly after three months of SCL wear.**
- **GCD reduction was associated with lens materials but not with lens wearing modality.**
- **No significant change was observed in epithelial cell metaplasia.**

This chapter is based on the following original article: Sapkota K, Franco S, Sampaio P, Lira M. Effects of three months of soft contact lens wear on conjunctival cytology. Clin Exp Opt 2015 in Press.

3B.1 Introduction

The popularity of contact lenses (CL) is increasing and it is estimated that 140 million people use this mode of refractive error correction in the world.[1] CL are also worn for therapeutic purposes in certain ocular surface diseases to relieve pain or improve prognosis,[2] and for aesthetic purpose to enhance the appearance.[3] Recent studies have shown a possible use of CL on continuous drug delivery into the eyes[4] and monitoring tear glucose level.[5] Because CL are worn directly on the ocular surface, they may lead to adverse effects on morphologic, metabolic, cytological, and immunologic state of the ocular surface[6] and these effects are significantly higher in extended lens wearers.[7] Dry eye is the most common complaint found in CL wearers.[8,9] Soft CL wear disrupts normal tear physiology by thinning and breaking up the tear film, interrupting tear film reformation and rupturing the lipid layer, increasing the evaporation rate.[10] CL wear also alters the secretion of aqueous, lipid and mucin components of the tear film as well as changes in their biochemistry.[11] Many researchers found a decrease in tear mucin level with CL wear.[12,13] It may change the morphology of conjunctival epithelial cells and/or the number of goblet cells.[14,15]

Conjunctival cytological examination reveals early, subclinical, cytotoxic effects attributable to CL wear as well as to the preservatives and chelating agents in soft CL care systems.[16] Conjunctival impression cytology (CIC) is a minimum invasive method to assess the ocular surface with no side effects.[17] CIC involves the collection of cells from the conjunctival surface with the help of a special type of filter paper by impression on the surface and examination of the superficial layer(s) of conjunctival epithelium with different types of staining. It can be done with or without anesthesia.[18] It can be used in wide range of techniques from simple light microscopic examinations to polymerase chain reactions. With optical microscopy, epithelial cell morphology, goblet cell density (GCD) and the presence or absence of any inflammatory cells can be examined.[19] Many recent studies conducted in conjunctival surface applying CIC found increased levels of epithelial squamous metaplasia and loss of goblet cell density in pathological conditions of the eyes.[17,18]

The majority of the previous studies concluded that CL wear reduces the number of goblet cells and increases epithelial cell metaplasia.[20-22] However, most of these studies were cross-sectional and they involved comparison of cytological data in CL wearers and with non-CL wearers. None of them evaluated the effect of the lens materials on conjunctival cytology.

Recently, many companies have introduced highly biocompatible lens materials which might behave differently on the ocular surface. The present longitudinal clinical trial was designed to investigate the changes in conjunctival cytology after three months of soft CL wear. GCD, as well as the changes in epithelial cell morphology, was determined. The effects of lens materials and wearing modality on the conjunctival cytology were also analyzed. Since, earlier studies found that the effects of CL wear on conjunctival cytology start within a few weeks, occurring rapidly during the initial period of lens wear and reaching maximum at about 3 months,[21,23,24] this trial was conducted for a period of three months.

3B.2 Methods

Study design

This was a longitudinal contra-lateral clinical trial conducted at University of Minho, Portugal. Ethical approval was obtained from Ethical Committee of the University of Minho. Each subject signed a consent form after the explanation of study procedures and its possible consequences and the tenets of Declaration of Helsinki were followed.

Subjects

Twenty-seven myopic subjects with normal ocular/systemic health, no previous history of CL wear or ocular surgery, and aged between 18 and 35 years were included in the study. Subjects with best corrected visual acuity less than 20/20 in one of the eyes, refractive astigmatism higher than 0.75D and pregnant women were excluded from the study.

Procedure

During the baseline visit, CIC was performed on the superior bulbar conjunctiva on both eyes of each subject.[18] Nitrocellulose Millipore[17] MF™-Membrane filter (MILLIPORE, Ireland) with pore size 0.45µm was used without application of topical anesthesia.[25,26] Briefly, a semi-circular piece of filter paper with diameter 13mm, touched the superior bulbar conjunctiva for few seconds and was removed in a gentle peeling motion (Figure 3B.1). The paper was then stained with PAS, Haemotoxylin and Eosin[27] and the slides were observed by an optical microscope with total magnification 100X and 400X. Goblet cells were counted in the higher power field (with a total magnification of 400X) and GCD was calculated as the number of cells per square millimetre.

Figure 3B.1. Photo during the specimen collection from the superior bulbar conjunctiva of a subject using Nitrocellulose Millipore MF™ Membrane filter. [Photo taken with written consent of the subject]

Morphological changes in epithelial cells including shape, size and characteristics of the nucleus were noted and graded according to Tseng classification [Table 3B.1].[28] This procedure was repeated in three random fields of area and the average was used in the analysis.

Table 3B.1. Conjunctival epithelial cell metaplasia classification[28]

Classification	Goblet cells	Epithelial cells	Nucleus cytoplasm ratio
Grade 0	Moderate density	Uniform size/form	1:1
Grade 1	Decreased density	Mild enlargement	1:2-1:3
Grade 2	Absent	Moderate enlargement, flattened (squamoid)	1:4
Grade 3	Absent	Markedly squamoid	1:6
Grade 4	Absent	Markedly squamoid, large	1:8
Grade 5	Absent	Shrunken cytoplasm	Nucleus may be absent

Lenses and solutions

Subjects were fitted with a daily disposable lens (Nelfilcon A or Stenofilcon A) in one eye and a monthly disposable (Lotrafilcon B or Comfilcon A) in the other eye. CL details are presented in table 3B.2. Post-lens fitting evaluation was performed and refitting with another type of CL among the study lenses was done where fitting was unacceptable. Subjects were well trained for CL usage, care and maintenance. In the dispensing time, CL, lens case and solution were provided for each subject for the coming month. Moreover, a paper with information about types and lens care methods was provided where they should indicate the number of wearing hours every day. This helped participants to wear the lenses correctly in the recommended eye. Subjects were informed to wear lenses at least 5 days in a week and a minimum of 8 hours per day, except during the first week, where the number of wearing hours per day was flexible. There was no limit on the number of days or hours when the lenses could be worn, however, they were not allowed to wear lenses during sleep, swimming or bathing. At the end of every month, participants should visit the office to monitor the adequacy of lens and solution use, and to provide new lens and lens care product for the next month.

For the first and second months, all the subjects used OPTI-FREE Puremoist solution (Polyquad 0.001% and Aldox 0.0006%, Alcon Laboratories, TX) while for the third month, 16 subjects used OPTI-FREE Puremoist and 11 subjects used AOSEPT PLUS (Hydrogen peroxide 3%, Alcon Laboratories, TX).These two types of solution were provided for each subject since another objective of the study was to investigate the effect of different types of solutions in CL wearers' comfort (to be published). There was no significant difference in GCD reduction and epithelial cell metaplasia between the eyes with these solutions ($p > 0.05$). Subjects were advised to contact the researcher at any time if they felt adverse events with the necessary management. CIC was repeated after three months of CL wear.

Statistical analysis

Data were analyzed with Statistical Package for Social Science (SPSS 22, IBM Corp., Armonk, NY). Descriptive statistics were expressed in mean ± standard deviation (SD). Kolmogorov-Smirnov test was performed to examine the normality of the variables. Parametric tests were applied for normally distributed variables while non-parametric tests were applied for others. One way analysis of variance with Post Hoc testing was applied to compare the reduction

in GCD between different lens materials. Spearman' rho was used to test the correlation of changes in GCD with the age of the subjects. Chi-Square test was applied to examine the proportion in changes in epithelial cell metaplasia. P values of < 0.05 were considered as statistically significant.

Table 3B-1. Characteristics of the lenses used in the study

	Lotrafilcon B	Nelfilcon A	Comfilcon A	Stenofilcon A
Company	Alcon	Alcon	Cooper Vision	Cooper Vision
Brand name	AirOptix® Aqua™	Dailies® AquaComfort Plus®	Biofinity™	MyDay™
Water content (%)	34	69	48	54
Thickness (mm)	0.08	0.10	0.08	0.07
Base curve /diameter (mm)	8.6/14.2	8.7/14	8.7/14.5	8.4/14.2
Oxygen Permeability (barrer)	110	26	128	80
Modulus (MPa)	1.2	0.89	0.75	0.4
Transmissibility (barrer/cm)	137.5	26	160	100

3B.3 Results

Fifty-four eyes of 27 subjects (63.0 % female) were included in this longitudinal clinical trial. The mean age of the subjects was 23.5±3.0 years (range 20-33 years). CL were worn as follows: Lotrafilcon B (n = 12), Nelfilcon A (n = 12), Comfilcon A (n = 15), Stenofilcon A (n = 15) and the mean power of the lenses used was -1.98 ± 1.60D (range -0.50 D to -5.50 D, 95 % CI, -1.60 D to -2.45 D). All subjects completed the study without any significant adverse event except for one case that showed a CL-induced peripheral ulcer in right eye wearing a Lotrafilcon B

lens. This complication was resolved after 10 days of lens wear discontinuation without any additional treatment and this subject continued the study.

Figure 3B.2 shows the representative images of the cytology with 100X total magnification. GCD was significantly reduced by 85 ± 151 cells/mm^2 ($p < 0.001$) [Figure 3B.3].

a.

b.

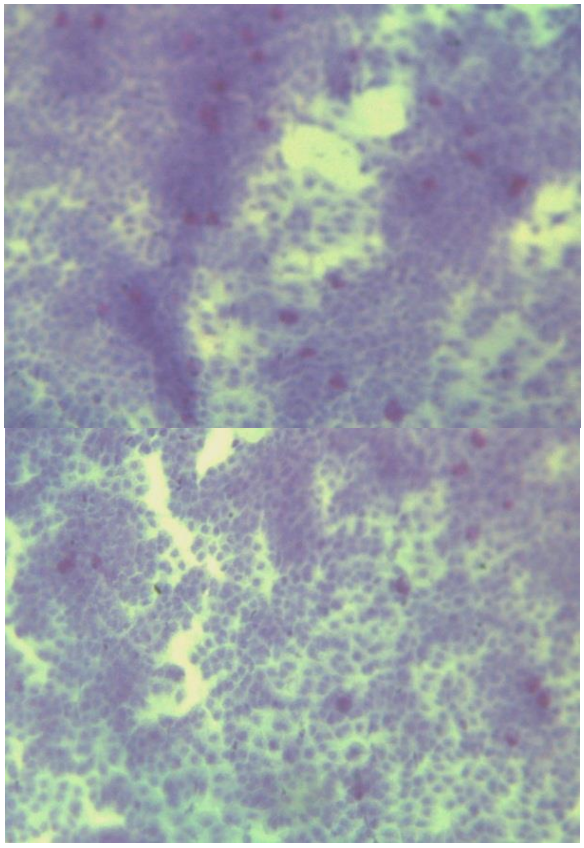


Figure 3B.2. Representative images of the conjunctival impression cytology (100X total magnification) of a subject: A. Before starting to wear contact lenses, B. After three months of contact lenses wear.

Figure 3B.4 shows the Bland-Altman graph showing the change in GCD with the initial GCD. Reduction in GCD was strongly correlated with the baseline GCD ($r = 0.846$, $p < 0.001$).

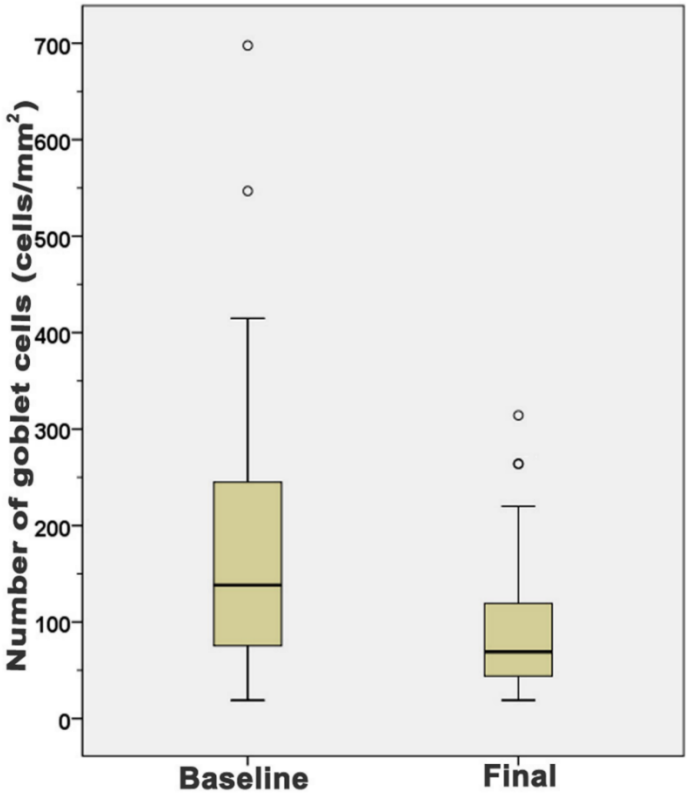


Figure 3B.3. Goblet cell density before (baseline) and after contact lens wear (final) [N = 54].

There was no significant difference in the baseline GCD among the eyes with different lenses ($p = 0.069$). As shown in table 3B.3, the magnitude of the reduction in GCD was significantly associated with CL materials ($p = 0.034$); the reduction was the greatest with Nelfilcon A lens wear (166 ± 147 cells/mm^2) while it was least with Comfilcon A lens wear (32 ± 137 cells/mm^2). Multiple comparisons showed that Nelfilcon A lens wear had a higher reduction than Comfilcon A and Stenofilcon A lens wear ($p < 0.05$). There was no significant difference between Nelfilcon A and Lotrafilcon B, between Lotrafilcon B and Comfilcon A or Stenofilcon A and between Comfilcon A and Stenofilcon A lens wear ($p > 0.05$). The reduction of GCD was not associated with the wearing modality of the lenses; it was similar with daily disposable and monthly disposable lenses ($p = 0.332$). There was no significant correlation between the decrease in GCD and age ($p = 0.160$) of the subjects.

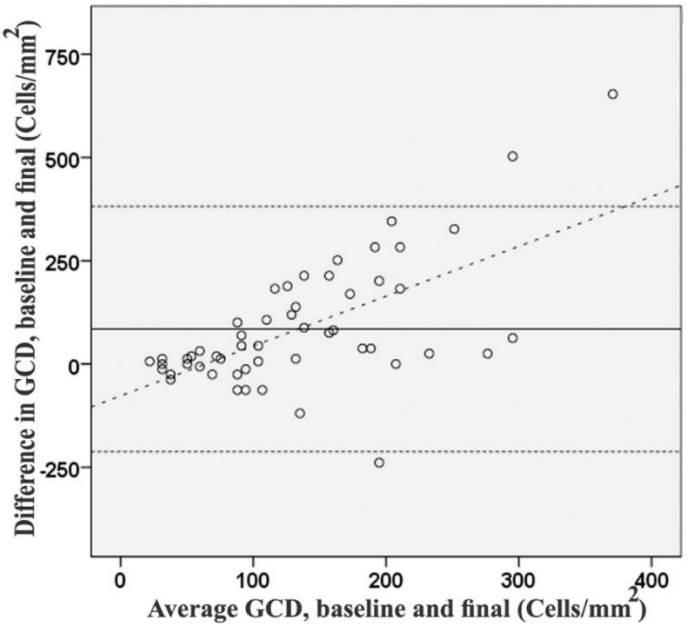


Figure 3B.4. Bland-Altman plotting showing the relation between changes in goblet cell density with the average goblet cell density [N = 54].

Table 3B.2. Reduction in goblet cell density with different types of study contact lenses

	Goblet cell density (Cells/mm^2)			
	Baseline	Final	Reduction	p values
Lotrafilcon B	239+164	132+79	107±187	0.018
Nelfilcon A	254+122	89+61	166±147	0.002
Comfilcon A	219+106	188+76	32±137	0.414
Stenofilcon A	219+108	164+30	53±113	0.362
Total (N = 54)	233+137	148+67	85±151	<0.001

During that period, no significant change in epithelial cell metaplasia was observed (p = 0.075). However, as shown in Table 3B.4, epithelial cell metaplasia grading increased in 74% of the eyes by at least one grade due to CL wear. The change in epithelial cell morphology was correlated with the age of the subjects (Spearman's rho = 0.286, p = 0.036). It was not associated with wearing modality of the lenses (p = 0.850). With silicone lens wear, 69.0% of the tested eyes

changed at least one grade while for non-silicone hydrogel lens wear the value found was 91.7%; however, changes in epithelial cell metaplasia were not significantly related to the lens material ($p = 0.158$).

Table 3B.3. Changes in conjunctival epithelial cell morphology after three months of soft contact lens wear

		Final			Total
		Grade 0	Grade 1	Grade 2	
Baseline	Grade 0	8 (19.0%)	27 (64.3%)	7 (16.7%)	42 (100.0%)
	Grade 1	0 (0.0%)	5 (45.5%)	6 (54.5%)	11 (100.0%)
	Grade 2	0 (0.0%)	1 (100.0%)	0 (0.0%)	1 (100.0%)
Total		8 (14.8%)	33 (61.1%)	13 (24.1%)	54 (100.0%)

3B.4 Discussion

In this study, the effects of three months of soft CL wear on conjunctival cytology was evaluated; subjects were neophyte CL wearers and different types of CL (one conventional hydrogel – Nelfilcon A, one silicone hydrogel lens with surface plasma treatment - Lotrafilcon B, one silicone lens without surface treatment – Comfilcon A and one new CL recently available in the market with smart silicone chemistry– Stenofilcon A) were used. Two of the lenses (Nelfilcon A, Stenofilcon A) are daily disposable lenses while the other two (Lotrafilcon B, Comfilcon A) are monthly disposable lenses. Lenses were worn in a contra-lateral manner in such a way that one eye was fitted with a daily disposable and the other with a monthly disposable lens.

In this study, we found a significant reduction in GCD after three months of CL wear. This reduction in GCD may explain the origins of CL-induced dry eye.[29] As depicted in Figure 3B.2, the initial GCD was highly varied and so was the changes in GCD. Consistent with our findings, Doughty using meta-analysis, found a high variation in GCD ranging between 10.5±1.1 and 152.85±29 cells/mm^2 in soft CL wearers.[20] Eyes with higher baseline GCD were found to suffer higher changes. The effect of CL wear on conjunctival GCD may be due to the physical and mechanical effect of the lens. Each blink induces CL movement as well as some friction of the

upper lid on the superior bulbar conjunctiva, the part which was used for CIC. Our results are consistent with studies that report a reduction in GCD induced by soft CL wear.[20] Simon *et al*. found a significant decrease in GCD following six-months of soft CL wear.[22] Knop and Brewitt found a decrease in GCD after 3-6 month of soft CL wear and the degradation of conjunctival cytology started within the first few weeks.[21] In another comparative study of conjunctival cytology in CL wearing subjects and non-CL wearing controls, Cakmak *et al*. found significant degradation of epithelial cell morphology and GCD.[30] Contrary to our findings, some studies found an increase in GCD after soft CL wear. Lievens *et al*. found an increase in GCD following six months wear of Acuvue 2 and PureVision CL.[31] Connor *et al*. also found a nearly two-fold increase in GCD after six months of soft CL wear.[32] These authors speculated that this increase in GCD may be due to an adaptive response of the ocular surface.

Hirji *et al*. suggested that lens care solution plays a role in the changes in conjunctival cytology associated with CL wear.[16] However, in the current study, the reduction in GCD was not associated with the wearing modality of the lenses (daily disposable versus monthly disposable). This suggests that there is not any link between GCD reduction and lens care solution.

As shown in Table 3B.3, the number of goblet cells changed differently when different lens materials are used: the reduction was maximal for the Nelfilcon A lens wearers ($p = 0.002$) followed by Lotrafilcon B (0.018) but there was no significant reduction in Comfilcon A ($p = 0.414$) and Stenofilcon A lens wearers (0.362). The Nelfilcon A lens, which was the non-silicone hydrogel lens used in the study, was the thickest lens with the highest water content among the CL studied. This material has the lowest oxygen permeability and a high Young's modulus (only less than Lotrafilcon B). On the other hand, Comfilcon A lens has high oxygen transmissibility but low Young's modulus. So low oxygen permeability, high Young's modulus and high center thickness might be important factors that can affect conjunctival goblet cells. Although, Lotrafilcon B has high oxygen permeability, it significantly reduced the GCD. It is the only silicone lens used in this study having plasma treatment on its surface. Besides oxygen permeability, surface treatment may also affect GCD. On multiple comparisons, only Nelfilcon A lens wear showed a difference in GCD reduction compared to Comfilcon A and Stenofilcon A lens wear. Lievens *et al*. found that non-silicone hydrogel lenses create more irritation than silicone lenses.[31] A higher reduction in GCD by Nelfilcon A lens wear may be due to the higher irritation on the ocular surface. It is

interesting to know that the lenses which were manufactured by the same company had similar effects on conjunctival cytology. This highlights the effect of lens characteristics and designs on conjunctival goblet cells. In contrary to the present findings, Simon *et al*. did not show statistical difference in cytological changes with different lens materials.[22] Similarly, Lievens *et al*. did not find any difference in GCD in Acuvue 2 or PureVision CL wearers.[31] The reduction in GCD associated with CL material in the present study might be due to the different characteristics of lens used (Table 3B.2).

There was not a significant correlation between the decrease in GCD with age of the subjects. Zhu *et al*. concluded that GCD does not change with age, but function of the goblet cells decreases with age.[33] This may be the case but testing of the function of the goblet cells was out of the scope of this study. Moreover, age-related variation may not take place over a narrow range of age featured in the current study.

There was no significant change in conjunctival epithelial cell metaplasia with CL wear. However, as shown in Table 3B.3, it was increased by few grades in the majority of the eyes. Before wearing CL, "grade 0" was observed in 78% of the eyes, which was found in only 15% of the eyes after CL wear. Epithelial metaplasia in CL wearers is supposed to be due to the mechanical influence of the lens on the ocular surface.[34] Increase in epithelial cell metaplasia may be one of the causes of CL-induced dry eye because increased severity of dry eye and conjunctival epithelial cell metaplasia are associated with one other.[35] Grade change did not reach a significant level probably due to the fact that the change was only a single grade in the majority of the eyes (61%). Epithelial metaplasia was not associated with lens wearing modality. Silicone lens wear was less associated with changes in epithelial metaplasia by comparison with non-silicone hydrogel lenses, which might be due to the higher oxygen permeability of silicone lenses. Both the aforementioned lens material used in our study had similar modulus of rigidity, and so the mechanical influence of lens on the ocular surface is supposed to be similar.[34]

Previous studies have found that aging does not alter the epithelial cell morphology.[33] However, a significant correlation of the change in epithelial metaplasia with age was observed in the current study, indicating that adult lens wearers are more susceptible to epithelial metaplasia.

An already compromised ocular surface due to age may be more susceptible to damage by CL wear.

Contrary to the findings of the current study, several studies have shown significant differences in conjunctival epithelial metaplasia in CL wearers and non-CL wearers. Tomatir *et al*. found significant differences in CIC with CL wear for a period of 6.9±2.6 months (range 4-12 months).[36] However, they used different lens material: soft hydroxyethylmethacrylate (vinyl pyrrolidone copolymer) lenses in 40 eyes, polymacon lenses in 70 eyes and rigid gas permeable (RGP) in 40 eyes. Similarly, Simon *et al*. found significant changes in epithelial squamous metaplasia after six-month soft CL wear.[22] They also found that the magnitude of changes was correlated with the duration of lens wear and significantly higher in symptomatic wearers compared to non-symptomatic wearers. However, in that study, the number of soft CL wearers was small since only 11 subjects completed the study. Moreover, the significant change in that study may be due to longer duration (6 months) in comparison to the present study. Doughty and Naase found significant differences in epithelial cell size in non-CL wearers and successful daily CL wearers (with duration of 4-9 years).[37] Munshi *et al*. found a significant increase on epithelial squamous metaplasia grade in subjects who wear soft or RGP lenses in comparison to the control group.[24] They did not find any association of epithelial squamous metaplasia with the duration of CL wear. In a recent cross-sectional study conducted in European women, Doughty found significant higher epithelial squamous metaplasia in soft CL wearers than those of non-lens wearers.[38] In the latter study, in addition to cell enlargement, a larger nucleus size was observed in CL wearers.

In the current study, a highly significant reduction in GCD after three months of soft CL wear was observed but there was non-significant change in conjunctival epithelial cell morphology. This suggests that CL wear probably affects the number of goblet cells before there are observable changes to epithelial cell morphology.

Consistent with the study by Simon *et al*., we did not find any snakelike chromatin due to soft CL wear.[22] Moreover, it was not encountered any Neutrophil or Lymphocyte cell in the sample in contrary to a study conducted by Hirji *et al*.[16] This may be due to the short duration of CL wear by the subjects in this study.

There are some limitations in this study. We used a small sampling area (high power field of view with 400X total magnification) to count the goblet cells and it is likely to make some error because of the variability in goblet cells.[39] However, observations were made in three random areas and the average was used for analysis to minimize this error.

From this study, it can be concluded that soft CL wear reduces GCD, which is dependent upon the lens materials, but may not significantly change the conjunctival epithelial cell morphology. Oxygen permeability, material strength (Young's modulus), surface treatment and thickness of the CL are important factors that can induce conjunctival cytological changes. CIC may help to detect early changes on CL wearers. However, as suggested by Munshi *et al.*, cytological changes which occur during early periods of CL wear may be due to the adaptive changes of ocular surface.[24] To confirm the findings of this study, a long-term, longitudinal study may be helpful.

References

1. Stapleton F, Keay L, Jalbert I, Cole N. The epidemiology of contact lens related infiltrates. *Optom. Vis. Sci.* 2007;84(4):257-72.

2. Ambroziak AM, Szaflik JP SJ. Therapeutic use of a silicone hydrogel contact lens in selected clinical cases. *Eye Contact Lens* 2004;30(1):63-7.

3. Zhang L, Chan O, Roy L, Barr JT. A meta-analysis of studies on cosmetically tinted soft contact lenses. *Clin. Ophthalmol.* 2013;7:2037-2042.

4. Garhwal R, Shady SF, Ellis EJ, Ellis JY, Leahy CD, McCarthy SP, Crawford KS GP. Sustained ocular delivery of ciprofloxacin using nanospheres and conventional contact lens materials. *Investig. Ophthalmology Vis. Sci.* 2012;53(3):1341-52.

5. Yao H, Shum AJ, Cowan M, Lähdesmäki I, Parviz BA. glucose level. *Biosens Bioelectron* 2011;26(7):3290-3296.

6. Stapleton F, Stretton S, Papas E, Skotnitsky C, Sweeney DF. Silicone Hydrogel Contact Lenses and the Ocular Surface. *Ocul. Surf.* 2006;4(1):24-43.

7. Efron N, Morgan PB, Hill EA, Raynor MK, Tullo AB. Incidence and morbidity of hospital-presenting corneal infiltrative events associated with contact lens wear. *Clin. Exp. Optom.* 2005;88(4):232-239.

8. Begley CG, Chalmers RL, Mitchell GL, *et al*. Characterization of ocular surface symptoms from optometric practices in North America. *Cornea* 2001;20(6):610-8.

9. Kastelan S, Lukenda A, Salopek-Rabatić J, Pavan J, Gotovac M. Dry eye symptoms and signs in long-term contact lens wearers. *Coll Antropol* 2013;37(Suppl 1):199-203.

10. Thai LC, Tomlinson A, Doane MG. Effect of contact lens materials on tear physiology. *Optom. Vis. Sci.* 2004;81(3):194-204.

11. Mann A, Tighe B. Contact lens interactions with the tear film. *Exp. Eye Res.* 2013;117:88-98.

12. Yasueda S, Yamakawa K, Nakanishi Y, Kinoshita M, Kakehi K. Decreased mucin concentrations in tear fluids of contact lens wearers. *J. Pharm. Biomed. Anal.* 2005;39(1-2):187-95.

13. Pisella PJ, Malet F, Lejeune S, et al. Ocular surface changes induced by contact lens wear. *Cornea* 2001;20(8):820-5.

14. Greiner J V, Covington HI, Korb DR, Allansmith MR. Conjunctiva in asymptomatic contact lens wearers. *Am. J. Ophthalmol.* 1978;86:403-413.

15. Greiner J V, Allansmith MR. Effect of contact lens wear on the conjunctival mucous system. *Ophthalmology* 1981;88:821-832.

16. Hirji N, Scott J, Sabel A. Conjunctival cytology in hard and soft contact lens wear. *Ophthalmic Physiol. Opt.* 1985;5(3):333-335.

17. Calonge M, Enrı A, Salamanca D, *et al*. Impression cytology of the ocular surface : a review. *Exp. Eye Res.* 2004;78:457-472.

18. Egbert PR, Lauber S MD. A simple conjunctival biopsy. *Am. J. Ophthalmol.* 1977;84(6):798-801.

19. Lopin E, Deveney-Tatiana B, Asbell P. Impression Cytology : Recent Advances and Applications in Dry Eye Disease. *Ocul. Surf.* 2009;7(2):93-110.

20. Doughty MJ. Contact lens wear and the goblet cells of the human conjunctiva — A review. *Contact Lens Anterior Eye* 2011;34(4):157-163.

21. Knop E, Brewitt H. Induction of conjunctival epithelial alterations by contact lens wearing. *Ger J Ophthalmol* 1992;1(3-4):125-34.

22. Simon P, Jaison SG, Chopra SK, Jacob S. Conjunctival Impression Cytology in Contact Lens Wearers. *Indian J Ophthalmol* 2002;50:301-306.

23. Doughty MJ. Contact lens wear and the development of squamous metaplasia of the surface cells of the conjunctiva. *Eye Contact Lens* 2011;37(5):274-81.

24. Munshi MM, Sathe V, Ganar A. Conjunctival impression cytology in contact lens wearers. *Cytopathology* 2001;12:314-320.

25. Blades K, Doughty MJ, Patel S. Pilot Study on the Use of Impression Cytology Specimens for Quantitative Assessment of the Surface Area of Bulbar Conjunctival Cells. *Optom. Vis. Sci.* 1998;75(8):591-9.

26. Thatcher RW, Darougar S JB. Conjunctival impression cytology. *Arch Ophthalmol* 1977;95(4):678-81.

27. Rivas L, Oroza MA, Perez-Esteban A M-CJ. Topographical distribution of ocular surface cells by the use of impression cytology. *Acta Ophthalmol* 1991;69(3):371-6.

28. Tseng S. Staging of conjunctival squamous metaplasia by impression cytology_ Tseng 1985. *Ophthalmology* 1985;92(6):728-33.

29. Shimazaki-Den S, Dogru M, Higa K, Shimazaki J. Symptoms, visual function, and mucin expression of eyes with tear film instability. *Cornea* 2013;32(9):1211-8.

30. Cakmak SS, Unlü MK, Karaca C, Nergiz Y, Ipek S. Effects of soft contact lenses on conjunctival surface. *Eye Contact Lens* 2003;29(4):230-3.

31. Lievens CW, Connor CG, Murphy H. Comparing goblet cell densities in patients wearing disposable hydrogel contact lenses versus silicone hydrogel contact lenses in an extended-wear modality. *Eye Contact Lens* 2003;29(4):241-4.

32. Connor CG, Campbell JB, Steel SA, Burke JH. The effects of daily wear contact lenses on goblet cell density. *J. Am. Optom. Assoc.* 1994;65(11):792-4.

33. Zhu W, Hong J, Zheng T, Le Q, Xu J, Sun X. Age-related changes of human conjunctiva on in vivo confocal microscopy. *Br. J. Ophthalmol.* 2010;94(11):1448-53.

34. Albietz J. Conjunctival histologic findings of dry eye and non-dry eye contact lens wearing subjects. *CLAO J* 2001;27(1):35-40.

35. Murube J, Rivas L. Impression cytology on conjunctiva and cornea in dry eye patients establishes a correlation between squamous metaplasia and dry eye clinical severity. *Eur J Ophthalmol* 2003;13(2):115-27.

36. Tomatir DK, Erda N, Gürlü VP. Effects of different contact lens materials and contact lens-wearing periods on conjunctival cytology in asymptomatic contact lens wearers. *Eye Contact Lens* 2008;34(3):166-8.

37. Doughty MJ, Naase T. Nucleus and cell size changes in human bulbar conjunctival cells after soft contact lens wear, as assessed by impression cytology. *Contact Lens Anterior Eye* 2008;31:131-140.

38. Doughty MJ. Objective Assessment of Squamous Metaplasia of Conjunctival Epithelial Cells as Associated With Soft Contact Lens Wear Versus Non-Lens Wearers. *Cornea* 2014;in press.

39. Doughty MJ. Sampling area selection for the assessment of goblet cell density from conjunctival impression cytology specimens. *Eye Contact Lens* 2012;38(2):122-9.

CHAPTER 4.

OCULAR SURFACE PHYSIOLOGY CHANGES WITH THREE MONTHS OF SOFT CONTACT LENS WEAR

Highlights

- **Changes in conjunctival bulbar and limbal redness and conjunctival and corneal staining due to three months of soft contact lens (SCL) wear was investigated.**
- **Conjunctival redness and conjunctival and corneal staining were increased.**
- **These changes were higher during initial period.**
- **Such changes were found to be associated with lens materials but not with wearing modality.**
- **Bulbar redness was found higher in lateral sides while corneal staining was found higher in inferior region.**

This chapter is based on the following original article: Sapkota K, Franco S, Lira M. Ocular surface physiology changes with three months of soft contact lens wear. Submitted to Optometry and Vision Science in October 2015.

4.1 Introduction

Contact lenses (CL) wear can induce metabolic, mechanical and toxic effects on the ocular surface.[1] Metabolic effect is considerably related with the oxygen transmissibility (Dk/t) of the lens materials.[2] A well-known study by Holden and Mentz found that a minimum of 24.1 unit of Dk/t is necessary to avoid the hypoxic effect on the ocular surface on daily wear.[3] Generally, manufacturers provide Dk/t of a minus three-diopter lens in the center region although Dk/t highly varies from center to periphery and from one power to the other.[4] So, Dk/t value given by manufacturer cannot represent the oxygen transmissibility of a whole lens. Moreover, for the ocular health, peripheral oxygen transmissibility of the lens is also important.[5] This shows some modern hyper permeable lenses may not be able to provide enough Dk/t to avoid hypoxic effect of lenses specifically in high power minus lenses on the peripheral region.[5] Mechanically, CL wear affects corneal as well as conjunctival health which depends upon the lens material characteristics.[6] Because soft CL are always kept with a liquid to protect from dehydration or for care and maintenance, the chemicals absorbed in lenses release on the ocular surface during wear inducing different toxic reactions.[7] Whatever may be the etiology of adverse effects of lens wear, it catalyze the inflammatory reaction which is initially observed with conjunctival redness due to the vasodilation and white blood cell migration.

A success in CL wear is determined by clear vision and comfort during the full-time wear. With the availability of many lens designs and parameters, clear vision is easily maintained with proper lens selection. However, due to its multifactorial etiology, comfort is always a challenge for CL wearers.[8] It is the main cause of CL discontinuation.[9] Comfort depends upon lens material characteristics, lens care products (LCP) as well as ocular surface health and also the environment.

Introduction of daily disposable lenses reduced comfort problems related to LCP use. However, recent studies found that daily disposable lenses are also not free of the effect of chemicals. Blister package solution also found to affect the ocular surface physiology.[7] Similarly, silicone lenses with hyperpermeability, reduced or eliminated hypoxia-related effect on the ocular surface.[10] Still inflammatory and mechanical side effect are not reduced but increased due to silicone hydrogel lenses.[11]

Ocular redness is the principal sign of eye inflammation. Generally limbal redness indicates corneal problems while diffused bulbar redness indicates conjunctival problems. The majority of the recent studies showed that ocular surface physiology is similar with and without modern lenses.[10,12] However, some studies pointed out that mechanical or inflammation related problems increased with silicone hydrogel CL wear.[13,14,15] Conjunctival physiology including bulbar redness, limbal redness and staining may be important factors for CL comfort; however, this area has not been extensively investigated.

The aims of this study were to determine the changes in conjunctival bulbar and limbal redness, and conjunctival and corneal staining due to three months of soft CL wear. The effect of lens material characteristics, wearing modality on the changes in ocular surface physiology was also investigated.

4.2 Methods

It was a longitudinal, contra-lateral study conducted in neophyte CL wearers in University of Minho, Portugal. Each subject signed a consent form after the study protocol was explained. Ethical approval was obtained from the Ethical Committee of School of Science of University of Minho. Study followed the tenets of Declaration of Helsinki.

Myopic subjects with astigmatism less than 1.00D who had never worn CL participated in the study. Subjects with previous history of ocular surgery, eye pathology and systemic disease were excluded from the study. Subjects presenting Efron grading scale equal or more than grade 2 were also excluded from the study. They should have commitment that they follow the protocol of the study. A sample size of at least 15 eyes was necessary to warrant the power of 0.99 to detect a difference of 0.5 unit in limbal or bulbar redness and conjunctival or corneal staining with p value of 0.05.[6]

Each subject wore a monthly disposable lens (Lotrafilcon B or Comfilcon A or Balafilcon A) in one eye and a daily disposable lens (Nelfilcon A or Stenofilcon A or Nesofilcon A) in the other eye, selected randomly. Lens details are presented in table 4.1. All the studied lenses were silicone hydrogel lenses except Nelfilcon A and Nesofilcon A. Monthly disposable lenses were

worn on daily wear basis that means lenses were removed during the night and disinfected in the provided solution and were replaced every month. Daily disposable lenses were discarded after single use. All the subjects were well taught about lens fitting and handling procedures. During the dispensing time, lenses and lens care product were provided for one month. They were told to come for the follow-up visit after one month when ocular examination was done and lenses and lens care product were provided for the next month. This process was continued until the third follow-up visit.

An information sheet was provided to each subject where they should note the number of CL wearing hours every day. Compliance with the protocol of the study was assured and reiterated in the subsequent visits. During the first week, number of wearing hours per day and number of wearing days per week were flexible; however, after the second week, all subjects were told to wear lenses at least 5 days per week and 8 hours per day.[16] There was not any limit of wearing period but lens wear during sleep and swimming were prohibited. During first two months, all subjects used OPTIFREE Puremoist multipurpose disinfecting solution (Polyquad 0.001% and Aldox 0.0006%, Alcon Laboratories, TX) while, during the third month, subjects used either OPTIFREE Puremoist or AOSEPT (Hydrogen peroxide 3%, Alcon Laboratories, TX) solution for the care of monthly lenses. This was done because other aim of the study (to be published) was to determine the effect of solution on lens wear comfort.

Slit lamp evaluation was performed on baseline visit, I month follow-up visit, II month follow-up visit and III month follow-up (final) visit. Conjunctival redness was observed in white light of slit lamp. Bulbar redness and limbal redness grading's were done in four regions: nasal, temporal, superior and inferior according to the Efron grading scale. Average values were used for the analysis. Conjunctival staining was observed in four regions after the application of Lissamine Green staining, and the average was used for analysis.[17] Corneal staining was observed with the application of 1% fluorescein and cobalt blue light filter and Wratten 12 barrier filter were used.[17] It was quantified in five areas: central, nasal, temporal, superior and inferior and average was used for the analysis.

Table 4-1. Characteristics of the contact lenses used in the study

	Lotrafilcon B	Comfilcon A	Balafilcon A	Stenofilcon A	Nelfilcon A	Nesofilcon A
Company	Alcon	Cooper Vision	Bausch & Lomb	Cooper Vision	Alcon	Bausch & Lomb
Brand name	AirOptix® Aqua™	Biofinity™	Purevision2™	MyDay™	Dailies® AquaComfort Plus®	Biotrue™ ONEday
Water content (%)	34	48	36	54	69	78
Thickness (mm)	0.08	0.08	0.07	0.08	0.10	0.1
Base curve/ diameter (mm)	8.6/14.2	8.7/14.5	8.6/14	8.4/14.2	8.7/14	8.6/14.2
Oxygen Permeability (barrer)	110	128	91	80	26	42
Modulus (MPa)	1.2	0.75	1.1-1.25	0.4	0.89	0.49
Transmissibility (barrer/cm)	137.5	160	130	100	26	42

Redness and staining were graded into 0 to 4 level in 0.1 increments; 0 representing normal and 4 representing the worst.[18] Examinations during each visit were done at least after 2 hours of wake-up to reduce the overnight physiological residual effect on the ocular surface. Similarly, to minimize the effect of diurnal variation on the ocular surface changes, every visit of each subject was scheduled for the same time of the day, for example, one patient whose baseline examination was done at 9-10:00 h, other follow-up examinations were also conducted in the same time and so on.[19]

Differential corneal staining, bulbar redness, limbal redness and conjunctival staining were calculated deducting the baseline values from the final values. These values in right and left eyes were not correlated so data from both the eyes were used in the analysis. Statistical Package for Social Sciences (SPSS 22) was used for the analysis of the data. Descriptive data were expressed in mean with standard deviation (SD). Kolmogorov-Smirnov test was applied to determine the normality of the data. Parametric tests and non-parametric tests were applied to detect the statistical relation in normally distributed and others variables respectively. Wilcoxon signed rank test was used to determine the changes in conjunctival bulbar /limbal redness, conjunctival staining and corneal staining before lens wear and after three months of lens wear. One way ANOVA was done for the multiple comparisons. P values less than 0.05 were assumed as statistically significant.

4.3 Results

Forty-seven myopic subjects participated in this study. The mean age of the subjects was 24.3+-4.1 years (range 19 to 35 years) and 66% (31) were females. Out of 94 eyes (mean refractive error -1.86±1.54D, range -0.50D to -5.60D), lenses were worn in the following way: Lotrafilcon B 16 eyes, Nelfilcon A 16 eyes, Comfilcon A 15 eyes, Stenofilcon A 15 eyes, Balafilcon A 16 eyes and Nesofilcon A 16 eyes. One way ANOVA showed no significant difference in baseline physiological signs and refractive error among different lens wearers ($p > 0.05$).

After three months of CL wear, bulbar redness increased significantly ($p < 0.001$). It was found to be increased during the first ($p < 0.001$) and the second month ($p < 0.001$), but there was no change in the third month ($p = 0.187$) in comparison to the values of the previous month. Bulbar

redness was found higher in the temporal and nasal side in comparison to superior and inferior regions ($p < 0.05$).

Limbal redness increased significantly ($p < 0.001$) after three months of CL wear. It increased during the first month ($p = 0.000$), second month ($p < 0.001$) and third month ($p = 0.028$). Limbal redness was found higher in temporal and nasal regions in comparison to superior and inferior regions ($p < 0.05$).

Corneal staining increased significantly after three months of CL wear ($p < 0.001$). It was increased during first month ($p = 0.000$), second month ($p = 0.000$) and third month ($p = 0.021$) in comparison to the values of previous month. Corneal staining was found significantly higher on the inferior corneal region in comparison to other regions ($p < 0.05$) and all the other regions had similar staining ($p > 0.05$).

Conjunctival staining increased significantly ($p < 0.001$) during every month in comparison to the values in the previous month. Figure 4.1 shows the increment observed in ocular surface physiology.

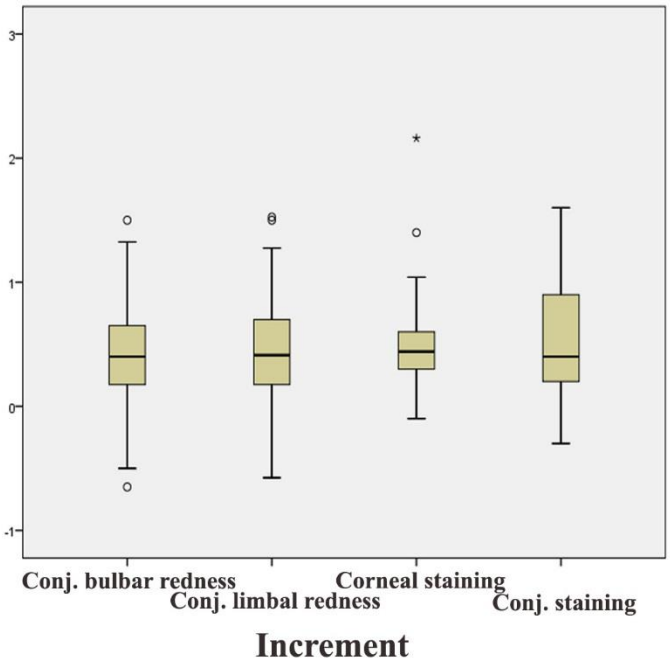


Figure 4.1. The increase in ocular surface physiology by three months of soft contact lens wear. [Conj. = Conjunctival]

No significant difference was observed in changes in conjunctival limbal and bulbar redness and corneal and conjunctival staining between daily and monthly disposable lenses ($p > 0.05$). There was a significant difference in bulbar redness ($p = 0.032$) and corneal staining ($p = 0.026$) between silicone and hydrogel lens wear but there was no significant difference in limbal redness and conjunctival staining ($p > 0.05$). Silicone lenses produced more bulbar redness and corneal staining than hydrogel lenses.

There was a correlation with lens power and the increase in bulbar redness ($r = -0.211$, $p = 0.043$) and the increase in limbal redness ($r = - 0.234$, $p = 0.025$) [higher power lenses produced higher changes] but it was not verified with corneal staining ($r = 0.005$, $p = 0.963$) and conjunctival staining ($r = -0.183$, $p = 0.083$). Table 4.2 shows the results obtained in ocular physiology for each lens separately.

Table 4-2. Increase in ocular surface physiology grading by three months of soft contact lens wear [Conj. = conjunctival]

Lens materials	Conj. bulbar redness (p)	Conj. limbal redness (p)	Corneal staining (p)	Conj. staining (p)
Lotrafilcon B	0.3 [0.001]	0.3 [0.002]	0.5 [0.001]	0.4 [0.001]
Comfilcon A	0.7 [0.001]	0.7 [0.001]	0.6 [0.001]	0.7 [0.001]
Balafilcon A	0.3 [0.008]	0.3 [0.009]	0.5 [0.001]	0.4 [0.001]
Nelfilcon A	0.4 [0.001]	0.4 [0.002]	0.4 [0.000]	0.4 [0.001]
Stenofilcon A	0.6 [0.001]	0.6 [0.001]	0.5 [0.001]	0.7 [0.002]
Nesofilcon A	0.3 [0.010]	0.3 [0.009]	0.3 [0.001]	0.4 [0.002]

On multiple comparison, increase in bulbar redness was found to be dependent upon material characteristics of the lenses ($p = 0.001$). Comfilcon A lens induced significantly higher bulbar redness than that of Lotrafilcon B ($P = 0.002$), Nelfilcon A lenses ($p = 0.010$), Balafilcon A ($p = 0.001$) and Nesofilcon A ($p = 0.000$) lenses. Stenofilcon A lenses induced more bulbar

redness than that of Lotrafilcon B ($p = 0.017$), Balafilcon A ($p = 0.010$) and Nesofilcon A ($p = 0.004$) lenses.

Increase in limbal redness was also found to be dependent upon the lens types and or designs ($p = 0.002$). Comfilcon A induced more limbal redness than that of Lotrafilcon B ($p = 0.001$), Nelfilcon A ($p = 0.010$), Balafilcon A ($p = 0.004$) and Nesofilcon A (0.001). Stenofilcon A induced more limbal redness than that of Lotrafilcon B ($p = 0.009$), Balafilcon A ($p = 0.025$) and Nesofilcon A ($p = 0.007$).

Increase in corneal staining was not associated with the lens types ($p = 0.233$). However, some lenses had significantly different amount of increase in corneal staining. Comfilcon A lenses induced more corneal staining than that of Nesofilcon A lenses ($p = 0.015$). Conjunctival staining was not associated with the lens materials ($p = 0.276$). All the lenses induced similar amount of conjunctival staining during three months of lens wear ($p > 0.05$).

During the third month, we used two types of solutions: OPTIFREE Puremoist for 21 subjects and AOSEPT for 24 subjects. All the physiological changes were higher with OPTIFREE Puremoist than AOSEPT, however, that was not statistically significant ($p > 0.05$).

4.4 Discussion

In this study physiological changes induced by three-month soft CL wear in forty-seven subjects were investigated. Moreover, effect of lens material characteristics and wearing modality was also studied. Change in one eye was compared with the change in the fellow eye in this contra-lateral study. Because, all the other related factors are similar between two eyes of each subject, this might provide the sole effect of lens wear.

Bulbar redness

The increase in bulbar redness was statistically significant after three months of CL wear. On the analysis of each month separately, it was seen that during the first two months, there was a higher change than during the third month where there was little or no change. CL wear affected temporal and nasal side heavily than the superior and inferior region which may be due to higher evaporation of tears on these exposed parts of the conjunctiva.[19] Bulbar redness related with CL wear may be due to mechanical irritation, lens movement, lens edge profile and modulus of

materials or solution toxicity. Glasson *et al*.[20] found an increase in bulbar redness due to short term CL wear. Carole *et al*. also found an increase in bulbar redness with soft CL wear after two weeks and 4 weeks period.[6] They did not mention about the variation of redness in different parts of the conjunctiva. However, they used different lens materials and the study duration was also different.

Limbal redness

Limbal redness can be an indicator of hypoxia due to lens wear and present study was observed an increase after three months of CL wear. It was increased every month but the change in the third month was smaller. Papas proposed that to avoid limbal redness, oxygen transmissibility on the peripheral lens should be more than 125 units.[21] Some of the lenses used in this study have central Dk/t superior to 125 units, however, none of them have peripheral Dk/t more than 125 as the Dk/t is significantly lower in the peripheral region of minus lenses than in the central region.[4] Present study registered higher changes in bulbar and limbal redness with higher power lenses. In agreement with the present findings, Carole *et al*. also found a similar increase in limbal redness after two weeks and after four weeks period of soft CL wear .[6] Similarly, Glasson *et al*.[20] found an increase in limbal redness with soft CL wear. Although it was statistically significant, clinicians should not concern with changes in limbal redness less than 1 grade in Efron grading scale found in this study. However, long term limbal redness can facilitate corneal neovascularization,[22] so practitioners should choose the lenses which induce minimal redness. The exposed regions of the conjunctiva, temporal and nasal parts, were the most affected parts and this may be due to higher tear evaporation in this region.[19]

Corneal staining

Corneal staining was also found to be increased during three months of CL wear. The p values in each month show that it was higher in initial two months and less in the third month. Contrary to our findings, Carole did not find significant changes in corneal staining after two weeks and four weeks of lens wear.[6] Jones *et al*. found corneal staining associated with lens and lens care product combination.[16] Corneal staining in soft CL wearers may also be due to loose or tight fitting of the lenses.[23] In the present study, subjects with unacceptably loose or tight fitting were excluded from the study. Consistent with a study conducted by Nichols *et al*., it was higher in the inferior corneal region in comparison to the other regions.[24] Higher staining on the inferior cornea may be related with incomplete blinking and drying of the inferior cornea.[25-,27] It may also be due to tear

film instability. In this study, we did not observe solution toxicity related corneal staining. Jones *et al*. purposed that, due to the higher thickness on the periphery in myopic CL, peripheral lens absorbs higher portion of the lens care product chemicals or blister package solution and it releases on the ocular surface. Peripheral corneal staining seems to be more common than in central cornea in solution toxicity related staining. In the current study, average corneal staining score was less than 1 which is clinically non-significant.[28] This may be because CL related corneal stainings are transient and can resolve after a short period.[29,30]

Conjunctival staining

Conjunctival staining has not been extensively studied, unlike corneal staining. In the current study, it was found a significant increase in conjunctival staining after CL wear. Conjunctival staining may be due to thin postlens tear film in the peripheral lens region around the limbus, lens material characteristics or lens design. Although conjunctival staining less than grade 2 in Efron grading is considered clinically non-significant,[31] it may affect the level of comfort in CL wearers. In consistent to our findings, Carole *et al*. also found significant conjunctival staining after two and four weeks of lens wear.[6]

Ocular surface physiology and CL wearing modality

Contrary to the findings of Jara *et al*.,[32] It was not observed significant difference in the changes in ocular surface physiology with wearing modality of the lenses since it was similar with daily disposable and monthly disposable lens wearers. They suggested that daily disposable lenses do not absorb lens care product and do not release the components on the ocular surface. In the present study, daily disposable lenses were worn after taking out directly from the blister package and it is possible that chemicals, like borate, present in the blister package solution may induce some allergic response in some patients.[7] Nichols *et al*. also did not find any association of wearing modality in corneal staining.[24]

Ocular surface physiology and CL materials

Bulbar redness and corneal staining were associated with the lens materials: with silicone hydrogel lenses, it was higher in comparison to the non-silicone hydrogel lenses. Bulbar redness and corneal staining may be dependent upon the material characteristics of the lenses like Young modulus, coefficient of friction, lens edge design rather than only Dk/t. It is known that oxygen

flux is more representative on oxygen supply to the ocular surface than Dk/t[33] and change in oxygen flux is very small after the increase in oxygen transmissibility from 25 units.[34] Limbal redness and conjunctival staining were similar with these materials. In contrast to our findings, Morgan *et al*[12] found less conjunctival staining with silicone lenses.

It was found that Comfilcon A lens wear induced higher limbal and bulbar redness than other lenses while Nesofilcon A induced least. Comfilcon A is the lens with highest oxygen permeability among the lenses used in this study. The larger diameter of the lens and higher Young modulus may be related with the conjunctival redness. This indicates that etiology of ocular surface physiology changes is multifactorial depending upon lens design, material characteristics, lens parameters rather than the oxygen permeability. From table 2, it is clear that, although oxygen permeability is medium and lens thickness is the highest among the studied lenses, Nesofilcon A induced minimum changes in all the physiological signs. Jara *et al*. also found different physiological changes with different lens materials.[32] Although there was not a significant difference in corneal and conjunctival staining between different lens materials, Comfilcon A induced more corneal staining than that of Nesofilcon A lens wear. Other interesting finding of this study was that all the changes were higher with Comfilcon A and Stenofilcon A lens wear (Table 4.2). Both of these lenses have the same centre thickness and are silicone hydrogel lenses manufactured by the same company and contain similar water content. This may indicate that lens manufacturing method and design may affect on ocular surface physiology.

Ocular surface physiology and CL power

It is interesting that bulbar redness and limbal redness changes were correlated with the dioptric power of lenses, higher the lens power, higher the changes. This may be due to the fact that higher power minus lenses have lower oxygen transmissibility because of increase in peripheral thickness.[4] Moreover, This may be due to the differential release of chemicals by the lenses on the ocular surface. Thicker peripheral part of the lenses absorbs more chemicals present in the lens care product and/or blister package solution and releases in the ocular surface. None of the previous studies investigated the role of refractive power of lenses on the conjunctival redness. The relation of lens power could not reach a significant level with corneal and conjunctival staining, however, Nichols found association of lens power with the corneal staining.[24]

Lens care products and ocular surface physiology

During the third month of CL wear, two types of lens care products were used to care the monthly lenses. There was not a significant difference in limbal and bulbar redness and conjunctival and corneal staining between subjects who used two types of lens care products. Interestingly, these physiological changes were minimum with peroxide-based lens care product in comparison to Polyquad based care product. It supports the previous study which found that peroxide care system is better in comparison to other multipurpose disinfecting system.[35] Jones *et al*. found the conjunctival redness changes dependent upon lens and lens care product.[16] They found an increase in limbal redness with Balafilcon A lens with polyaminopropyl biguanide solution but not with Polyquad based lens care product.

This study used a quantitative but somewhat subjective type of grading which may lead to some human error or bias in comparison to the objective grading.[36] However, we have tried to minimize such bias and in the current study average values of physiological changes were used in the analysis. However, it is known that it does not emphasize on clinical importance.[25] For example when inferior corneal staining score is grade 4 and the other regions are grade 0, the average would be 0.8, which is clinically insignificant. So, it is recommended to use the highest score for the analysis in the future studies.

In conclusion, three months of soft CL wear statistically significantly increased conjunctival redness and conjunctival and corneal staining. This effect was found maximum with Comfilcon A lens wear and minimum with Nesofilcon A lens wear. Ocular physiological changes due to lens wear were minimum with peroxide-based lens care product. Although all these changes were statistically significant, the mean values of each parameter were less than 1 grade which may be considered to be clinically non-significant.[28] However, symptomatic patients were found to have higher conjunctival and corneal signs.[32] Therefore, such minor problems may affect CL comfort and finally on the success of lens wear. CL practitioners are suggested to recommend the lenses which induce fewer changes on ocular surface physiology.

References

1. Guillon M, Shah D. Objective Measurement of Contact Lens Induced Conjunctival Redness. *Optom. Vis. Sci.* 1996;73(9):595-605.

2. Jacob JT. Biocompatibility in the development of silicone-hydrogel lenses. *Eye Contact Lens* 2013;39(1):13-9.

3. Holden BA, Mertz GW. Critical oxygen levels to avoid corneal edema for daily and extended wear contact lenses. *Invest. Ophthalmol. Vis. Sci.* 1984;25(10):1161-7.

4. Lira M, Pereira C, Real Oliveira MECD, Castanheira EMS. Importance of contact lens power and thickness in oxygen transmissibility. *Contact Lens Anterior Eye* 2015;38(2):120-126.

5. Papas EB. The significance of oxygen during contact lens wear. *Contact Lens Anterior Eye* 2014;37(6):394-404.

6. Maldonado-Codina C, Morgan PB, Schnider CM, Efron N. Short-term physiologic response in neophyte subjects fitted with hydrogel and silicone hydrogel contact lenses. *Optom. Vis. Sci.* 2004;81(12):911-921.

7. Peterson RC, Fonn D, Woods CA, Jones L. Impact of a rub and rinse on solution-induced corneal staining. *Optom. Vis. Sci.* 2010;87(12):1030-1036.

8. Guillon M. Are silicone hydrogel contact lenses more comfortable than hydrogel contact lenses? *Eye Contact Lens* 2013;39(1):86-92.

9. Dumbleton K, Caffery B, Dogru M, *et al*. The TFOS International Workshop on Contact Lens Discomfort: report of the subcommittee on epidemiology. *Invest. Ophthalmol. Vis. Sci.* 2013;54(11):TFOS20-36.

10. Sweeney DF. Have silicone hydrogel lenses eliminated hypoxia? *Eye Contact Lens* 2013;39(1):53-60.

11. Szczotka-Flynn L, Chalmers R. Incidence and Epidemiologic Associations of Corneal Infiltrates With Silicone Hydrogel Contact Lenses. *Eye Contact Lens* 2013;39(1):47-51.

12. Morgan PB, Chamberlain P, Moody K, Maldonado-Codina C. Ocular physiology and comfort in neophyte subjects fitted with daily disposable silicone hydrogel contact lenses. *Cont. Lens Anterior Eye* 2013;36(3):118-25.

13. Nichols JJ. Deposition on silicone hydrogel lenses. *Eye Contact Lens* 2013;39(1):20-23.

14. Lin MC, Yeh TN. Mechanical complications induced by silicone hydrogel contact lenses. *Eye Contact Lens* 2013;39(1):115-24.

15. Keir N, Jones LW. Wettability and Silicone Hydrogel Lenses : A Review. *Eye Contact Lens* 2013;39(1):100-108.

16. Jones LW, Macdougall N, Sorbara GL. Asymptomatic Corneal Staining Associated with the Use of Balafilcon Silicone-Hydrogel Contact Lenses Disinfected with a Polyaminopropyl. *Optom. Vis. Sci.* 2002;79(12):753-761.

17. Korb DR, Herman JP, Finnemore VM, Exford JM, Blackie C a. An evaluation of the efficacy of fluorescein, rose bengal, lissamine green, and a new dye mixture for ocular surface staining. *Eye Contact Lens* 2008;34(1):61-64.

18. Nathon E. Efron Grading Scales for Contact Lens Complications. In: Efron N, Ed. Contact Lens Complications. Oxford: Butterworth- Heinnemann; 2004:239-43.

19. Duench S, Simpson T, Jones LW, Flanagan JG, Fonn D. Assessment of variation in bulbar conjunctival redness, temperature, and blood flow. *Optom. Vis. Sci.* 2007;84(6):511-516.

20. Glasson MJ, Stapleton F, Keay L, Willcox MDP. The effect of short term contact lens wear on the tear film and ocular surface characteristics of tolerant and intolerant wearers. *Contact Lens Anterior Eye* 2006;29(1):41-47.

21. Papas E. On the relationship between soft contact lens oxygen transmissibility and induced limbal hyperaemia. *Exp. Eye Res.* 1998;67(2):125-131.

22. Junghans BM, Collin HB. The limbal vascular response to corneal injury. An autoradiographic study. *Cornea* 1989;8(2):141-149.

23. Young G, Coleman S. Poorly fitting soft lenses affect ocular integrity. *CLAO J* 2001;27(2):68-74.

24. Nichols KK, Mitchell GL, Simon KM, Chivers D, Edrington TB. Corneal staining in hydrogel lens wearers. *Optom. Vis. Sci.* 2002;79(1):20-30.

25. Carolyn G Begley, Joseph T Barr, Timothy B Edrington, William D Long, Curtis D McKenney RLC. Characteristics_of_Corneal_Staining_in_Hydrogel.12.pdf. *Optom. Vis. Sci.* 1996;73(3):193-200.

26. Schwallie J, Mckenney C, William L, Angela M. Corneal Staining Patterns in Normal Non Contact Lens Wearers.pdf. *Optom. Vis. Sci.* 1997;74:92-98.

27. Bandamwar KL, Garrett Q, Cheung D, *et al*. Onset time course of solution induced corneal staining. *Contact Lens Anterior Eye* 2010;33(4):199-201.

28. Pritchard N, Young G, Coleman S, Hunt C. Subjective and objective measures of corneal staining related to multipurpose care systems. *Contact Lens Anterior Eye* 2003;26(1):3-9.

29. Luensmann D, Moezzi A, Peterson RC, Woods C, Fonn D. Corneal Staining and Cell Shedding during the Development of Solution-Induced Corneal Staining. *Optom. Vis. Sci.* 2012;89(6):868-874.

30. Tran N, Graham AD, Lin MC. Ethnic differences in dry eye symptoms: Effects of corneal staining and length of contact lens wear. *Contact Lens Anterior Eye* 2013;36(6):281-288.

31. Guillon M, Maissa C. Bulbar conjunctival staining in contact lens wearers and non lens wearers and its association with symptomatology. *Contact Lens Anterior Eye* 2005;28(2):67-73.

32. De la Jara PL, Papas E, Diec J, Naduvilath T, Willcox MDP, Holden BA. Effect of Lens Care Systems on the Clinical Performance of a Contact Lens. *Optom. Vis. Sci.* 2013;90(4):344-350.

33. Fatt I. New physiological paradigms to assess the effect of lens oxygen transmissibility on corneal health. *CLAO J.* 1996;22(1):25-29.

34. Compañ V, López-Alemany A, Riande E, Refojo MF. Biological oxygen apparent transmissibility of hydrogel contact lenses with and without organosilicon moieties. *Biomaterials* 2004;25(2):359-365.

35. Keir N, Woods CA, Dumbleton K, Jones L. Clinical performance of different care systems with silicone hydrogel contact lenses. *Contact Lens Anterior Eye* 2010;33(4):189-195.

36. Sorbara L, Simpson T, Duench S, Schulze M, Fonn D. Comparison of an objective method of measuring bulbar redness to the use of traditional grading scales. *Contact Lens Anterior Eye* 2007;30(1):53-59.

CHAPTER 5.

EFFECT OF SOFT CONTACT LENS WEAR ON CORNEAL BIOMECHANICAL PROPERTIES

Highlights

- **Corneal resistance factor (CRF) and corneal hysteresis (CH) were measured with ocular response analyzer before and after soft contact lens (SCL) wear.**
- **CRF reduced significantly after three months of SCL wear.**
- **Reduction in CRF was associated with lens materials but not with lens wear modality.**
- **No change was obtained in CH.**

This chapter is based on the following original article: Sapkota K, Franco S, Lira M. Effect of soft contact lens wear on corneal biomechanical properties. Submitted to Clin Exp Optom in October 2015.

5.1 Introduction

The study of corneal biomechanical properties is always attracting many researchers because of the importance of these properties in the measurement/variation in of intraocular pressure (IOP),[1] in the analysis of its effect on glaucoma,[2,3] in corneal diseases like keratoconus[4,5] or Fuchs dystrophy and also to evaluate the efficacy of corneal refractive therapy lenses.[6] After the introduction of the ocular response analyzer (ORA, Reichert Ophthalmic Instruments, Buffalo, NY, USA), it is possible to measure in vivo the corneal biomechanical properties.[7] It measures corneal resistance factor (CRF) and corneal hysteresis (CH) including intraocular pressure in the same time. Details of this instrument can be found in earlier studies.[8,9] Briefly, it applies the principle of non-contact tonometry. A metered rapid air pulse applies pressure on the central cornea, so that cornea moves inward and then comes outward on its initial position when the air impulse ends. So, it makes inward and outward applanation, the whole process of which is monitored by an electro-optical system of the instrument. Thus, from these data of about 20 milliseconds, ORA acquires CRF and CH by quantifying the differential inward and outward applanation pressures as follows: CRF = P1-KP2 and CH = P1-P2; where P1 and P2 are inward and outward applanation pressures respectively and K is a constant.[10] CRF represents the corneal resistance to deformity as a measure of visco-elastic properties of the cornea. It predominantly relates to the elastic properties of the cornea and is supposed to be a linear function of inward and outward applanation pressures. CH is a measure of the corneal viscous damping and is supposed to be the effect of energy absorption by the cornea. Measurement of CRF and CH with ORA were found with high repeatability and reproducibility.[11]

Corneal biomechanical properties were found to be low in eyes with keratoconus and correlated with its degree.[7,12,13] Similarly, both myopic and hypermetropic refractive surgeries were found to reduce CH and CRF.[9,14] Beshtawi *et al*. reviewed corneal biomechanical properties data and concluded that ultraviolet A riboflavin crosslinking does not significantly change the biomechanical properties of the cornea.[15]

A handful of studies investigated short-term effect of specialty CL on corneal biomechanical properties and found controversial results.[16,17] However, as far as the knowledge of the authors, no studies have been conducted to determine the effect of soft CL wear on CRF and

CH. The aim of this study was to investigate the effect of soft CL wear on corneal biomechanical properties. CRF and CH were evaluated during the baseline visit and after three months of lens wear. The effect of lens materials on the changes was also determined. The change in Rc and its role on corneal biomechanical properties was also studied.

5.2 Methods

This was a longitudinal clinical trial conducted at University of Minho, Portugal. This study followed the Tenets of Declaration of Helsinki and ethical approval was obtained from Ethical Committee of School of Science of the university. All subjects were clarified about the research protocol and signed a consent form before starting the trial. A sample size of at least 15 subjects in each group was necessary to warrant a 0.8 statistical power in order to identify a difference in CRF or CH of 0.8 mmHg with alpha of 0.05.

Subjects were recruited from the university students who had never worn CL. Subjects were older than 18 years, had myopic refractive error with astigmatism $\leq$ 0.75D and had best-corrected visual acuity of 20/20 or better in each eye. Any participants with active ocular pathology, past ocular surgery, systemic disorder which affect the successful CL wear were excluded from the study. Subjects with Efron's grading 2 or more were also excluded.[18] A detailed ocular history, as well as a slit lamp examination, was done and subjects were selected based on the inclusion/exclusion criteria.

Subjects were fitted with a daily disposable lens (Nelfilcon A or Stenofilcon A or Nesofilcon A) in one eye and a monthly disposable lens (Lotrafilcon B or Comfilcon A or Balafilcon A) in the other eye. Detailed lens characteristics are shown in table 5.1. Daily disposable lenses were discarded after single use while monthly lenses were worn on daily wear modality and they were cleaned, soaked and disinfected with a multipurpose solution overnight and reused and discarded after one month. Subjects were well trained about insertion and removal of the lenses. On the dispensing visit, lens and lens care product were provided for the preceding month. A sheet of paper with information about proper lens usage was provided where subjects should note the number of wearing hours every day. They were called for a follow-up visit after one month. During the follow-up visit, an ocular examination was done besides the monitoring of the compliance on

lens care and maintenance. Again, lens and lens care product were provided for the coming month. This process was repeated until the completion of three months of CL wear when the final examination was performed. During the first and second month, each subject was provided with OPTIFREE Puremoist (Polyquad 0.001% and Aldox 0.0006%, Alcon Laboratories, TX) solution for the care of monthly disposable lenses while for the third month, either OPTIFREE Puremoist or AOSEPT Plus (Hydrogen peroxide 3%, Alcon Laboratories, TX) was provided randomly. During the final month, these two types of solutions were used because one other objective of the study was to determine the effect of different solutions on comfort.

CRF and CH of each eye of every subject were measured with ORA. Subject was seated in a comfortable chair with forehead on the forehead rest of the instrument and three readings of CRF and CH were obtained in each case; the average was used for the analysis. To ensure good quality of measurements, only readings with well-defined applanation peaks, symmetrical and regular in height were considered. Corneal curvatures were measured by Medmont E-300 (Medmont Pvt. Ltd., Australia) and the average of steep and flat was used for the analysis. All the tests were repeated during each monthly follow-up visit. To reduce the potential effect of diurnal variation in corneal biomechanical properties, follow-up visits were arranged on the same hour of the day as that of baseline visit.[19,20]

Data were analyzed with IBM SPSS 22 statistical software (IBM Corp., Armonk, NY). Descriptive variables were expressed in mean ± standard deviation (SD). Kolmogorov-Smirnov test was used to determine the distribution of the data, parametric tests were used in normally distributed variables and non-parametric tests were used in the others. One way analysis of variance with Post Hoc Test was performed to compare the variables with different lens groups. Wilcoxon Signed Rank tests were used to find out the statistical significance in change in CRF, CH and Rc before and after lens wear. Mann-Whitney test was performed to determine the association of corneal biomechanical variables with gender. Spearman's rho values were calculated to find out the correlation of change in corneal biomechanical properties with other variables like baseline corneal biomechanical properties and lens power. Bland-Altman plotting was done to determine the trend in changing CRF, CH and Rc with lens wear. P value less than 0.05 was considered as statistically significant. We included data of both the eyes as there was no

significant correlation in the changes in corneal biomechanical properties in between right and left eyes ($p > 0.05$).

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Table 5-1. Characteristics of the contact lenses used in the study

	Lotrafilcon B	Nelfilcon A	Comfilcon A	Stenofilcon A	Balafilcon A	Nesofilcon A
Company	Alcon	Alcon	Cooper Vision	Cooper Vision	Bausch & Lomb	Bausch & Lomb
Brand name	AirOptix® Aqua™	Dailies® AquaComfort Plus®	Biofinity™	MyDay™	Purevision2™	Biotrue™ ONEday
Water content (%)	34	69	48	54	36	78
Thickness (mm)	0.08	0.10	0.08	0.08	0.07	0.1
Base curve/ diameter (mm)	8.6/14.2	8.7/14	8.7/14.5	8.4/14.2	8.6/14	8.6/14.2
Oxygen Permeability (barrer)	110	27	128	80	91	42
Modulus (MPa)	1.2	0.89	0.75	0.4	1.1-1.25	0.49
Transmisibility (barrer/cm)	137.5	26	160	100	130	42

5.3 Results

Forty-six subjects were included in this study with mean age 24.4±4.1 years (range 19 to 35). About two-thirds (65.2%) were females.

Among the 92 eyes (mean CL power: -1.87±1.56 D), 16 were fitted with Lotrafilcon B, 15 with Comfilcon A, 15 with Balafilcon A, 16 with Nelfilcon A, 15 with Stenofilcon A and 15 with Nesofilcon A. There was no significant difference in CH, CRF and Rc at the baseline visit among the eyes in different lens wearing group ($p > 0.05$). CRF, CH and Rc values at the different evaluation visits are presented in figure 5.1.

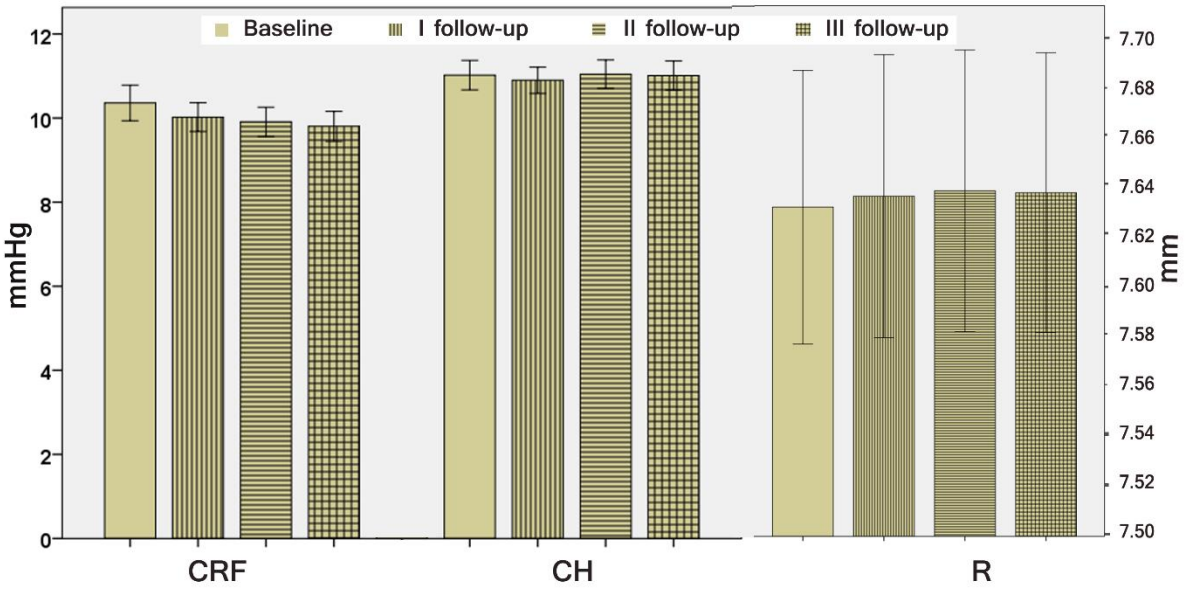


Figure 5.1. Mean CRF, CH and R values at the study visits, N = 92 [CRF: corneal resistance factor, CH: corneal hysteresis, Rc: average anterior corneal curvature].

As shown in table 5.2, there was statistically significant reduction in CRF after lens wear ($p < 0.001$) but there was no significant change in CH and Rc ($p > 0.05$). The reduction of CRF was correlated with baseline CRF (Spearman's Rho, $r = 0.457$, $p = < 0.001$) and baseline CH ($r = 0.378$, $p = < 0.001$) but not correlated with corneal curvature, CL refractive power, number of wearing hours and lens wearing modality ($p > 0.05$). Corneal flattening was not correlated with CRF and CH ($P > 0.05$). Bland-Altman graph [Figure 5.2] shows the trend of change in CRF, CH

and anterior corneal curvature with SCL wear. All the changes were within the first standard deviation of the mean changes. One way ANOVA shows that the changes in CRF is dependent upon the lens materials (p = 0.040) but there was no association of lens material with changes in CH and R (p >0.05).

Table 5-2. Changes in corneal biomechanical parameters with soft contact lens wear (N = 92).

	Baseline	Final	Changes	P values
CRF (mmHg)	10.24±1.91	9.76±1.58	0.48±1.15	0.000**
CH (mmHg)	10.90±1.60	10.96±1.55	-0.06±1.19	0.341**
Rc (mm)	7.64±0.25	7.65±0.26	-0.01±0.06	0.249*

CRF: corneal resistance factor, CH: corneal hysteresis, Rc: anterior corneal curvature, *Pair Sample T test, **Wilcoxon Signed Rank Test

On multiple comparisons by post-hoc test, the change was higher with Comfilcon A than with Balafilcon A (p = 0.014) and with Nesofilcon A (p = 0.003). Similarly, Stenofilcon A reduced CRF more than that of Nesofilcon A lens (p = 0.034) [Table 5.3]. The flattening of the corneal surface was less with Comfilcon A than with Lotrafilcon B (p=0.040), Nelfilcon A (p = 0.026) and Balafilcon A (p = 0.034).

Table 5-3. Change in biomechanical characteristics with different lens materials.

	ΔCRF	p*	ΔCH	p*	ΔR	p*
Lotrafilcon B	0.43±1.02	0.187	0.03±0.99	0.959	0.02±0.06	0.300
Comfilcon A	**1.13±0.94**	**0.000**	0.01±0.85	0.842	-0.03±0.08	0.638
Balafilcon A	0.12±1.72	0.460	0.05±1.97	0.776	**0.02±0.02**	**0.011**
Nelfilcon A	**0.58±0.96**	**0.039**	0.08±1.07	0.877	0.02±0.05	0.079
Stenofilcon A	**0.75±1.06**	**0.008**	0.27±1.09	0.460	0.002±0.09	0.972
Nesofilcon A	0.12±0.63	0.551	0.30±0.99	0.211	0.01±0.04	0.363

CRF: corneal resistance factor, CH: corneal hysteresis, Rc: anterior corneal curvature, *Wilcoxon Signed Ranks Test

Figure 5.2 shows the trend of change on CRF, CH and anterior corneal curvature with soft CL wear.

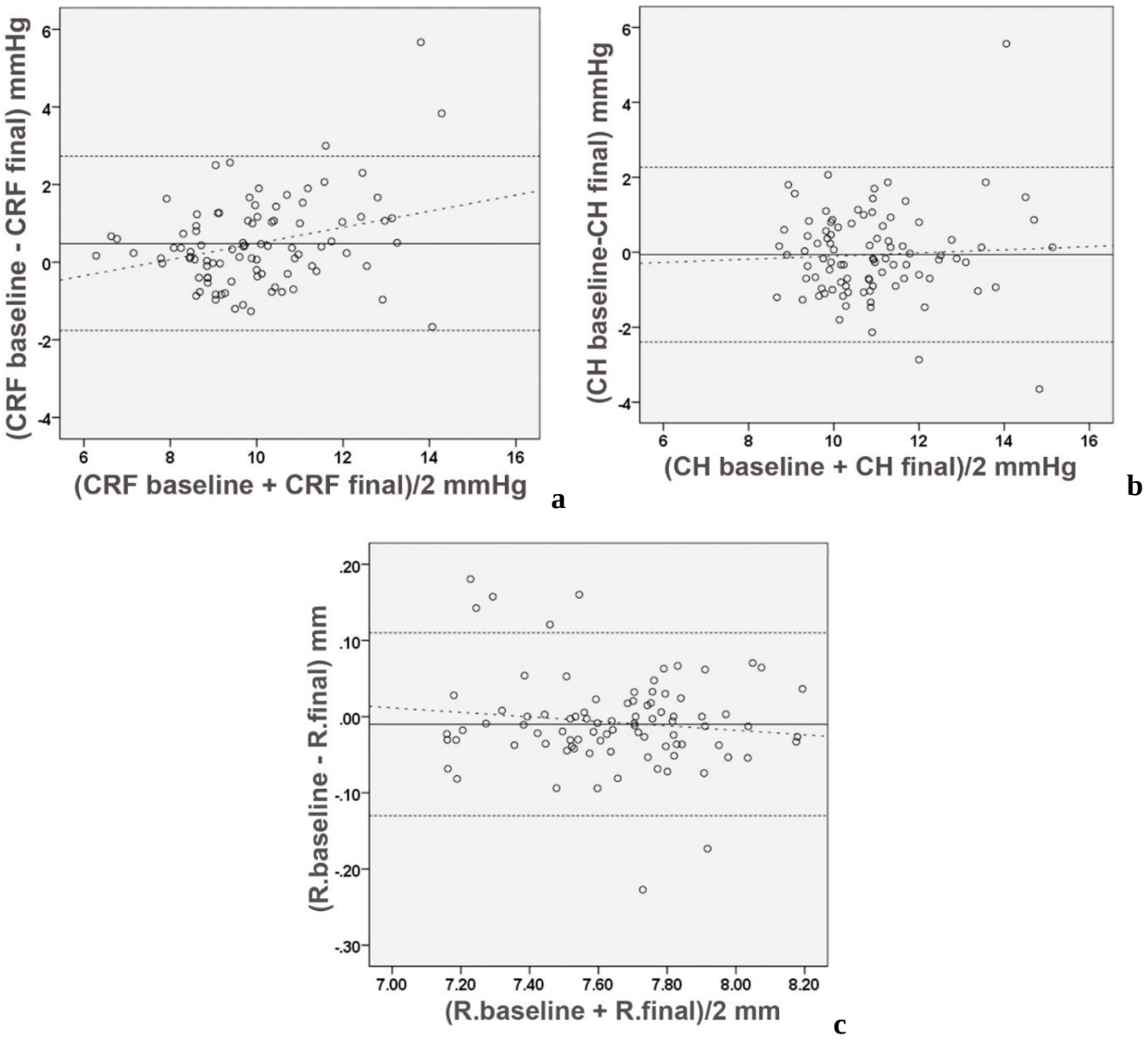


Figure 5.2. Bland-Altman graph showing the trend of change in a. CRF, b. CH, and c. Rc [CRF: corneal resistance factor; CH: corneal hysteresis; Rc: mean of the anterior corneal curvature].

5.4 Discussion

In this study, the effect of soft CL, silicone hydrogel and non-silicone hydrogel, on the corneal biomechanical properties were evaluated. The baseline values of both CRF and CH were found in a normal range as found in a previous study.[16]

It was found a reduction in CRF after three months of soft CL wear, however, there was no change in CH during this period. The cause of the reduction in CRF with soft CL wear is unclear. Supporting previous studies, no significant correlation was found between corneal curvature and CRF.[21,6] Narayanaswamy *et al*. suggested that CRF will be lower in flatter corneas.[22] In the current study, although it was not statistically significant, there was 0.01mm flattening of the cornea by CL wear. So this may be one cause behind the reduction in CRF. It was found that CRF is low in a weak cornea, so, reduction in CRF may indicate that soft CL wear makes the cornea fragile.[10] Lu *et al*.[23] induced corneal edema by using thicker CL on eye closure and measured corneal biomechanical properties by ORA. They found no change in CH with CL but an increase in CRF. They also suggested that CH is relatively stable with corneal thickness while CRF is positively correlated with corneal thickness. Franco and Lira also found a strong correlation of CRF with corneal thickness and weak correlation between CH and corneal thickness.[21] So, CH is expected to be same with CL wear while CRF might be increased due to some corneal edema (thickness). However, the finding in the current study was in contrary and this may be due to the fact that all the subjects wore lenses on daily wear basis and the oxygen transmissibility of these lenses were more than 24 unit which is sufficient to avoid corneal edema.[24]

Chen *et al*. found a decrease in CRF but no change in CH by short term orthokeratology lens wear.[16] They have suggested that the decrease in CRF may be due to change in the shape of the cornea rather than the change in corneal thickness. However, they have used corneal refractive therapy lenses with rigid materials, so the findings of that study could not be comparable with our study. Moreover, studies show that there is no correlation with corneal biomechanical properties and anterior corneal curvature.[21]

Nieto-Bona *et al*. found significant reduction in CH but non-significant decrease in CRF due to one month wear of corneal reshaping therapy (CRT) CL.[17] In the same study, they observed that after ceasing lens wear, both CH and CRF tend to increase although statistically non-

significant. They concluded that changes in corneal biomechanical properties due to CRT lens wear is a reversible process.[17] Gonjalez-Meijome *et al*. suggested that the effect, as well as recovery of orthokeratology therapy, correlates with corneal biomechanical properties.[6] They observed that effect of orthokeratology was higher with cornea presenting lower CH and CRF.

Reduction of CRF was found correlated with baseline CRF and CH. Touboul *et al*. suggested that a weak cornea has lower CH and CRF values.[10] So, it can be that with a strong cornea, there is greater reduction in CRF. It is interesting that reduction in CRF was not associated with lens power, number of wearing hours and type of lens. None of the subject's worn lenses overnight, but CRF reduction was independent upon the daily disposable or monthly disposable modality.

Narayanaswamy *et al*.[22] found better corneal biomechanical properties in women than in men. In contrary to that, in the current study, no difference in corneal biomechanical properties either in baseline and final or in the changes was obtained between men and women. Moreover, there was no statistically significant difference in corneal biomechanical properties in those females who were taking oral contraceptives and those who were not taking although Goldich *et al*. revealed that CRF and CH vary during ovulation cycle in women.[25]

The important finding of this study is that changes in CRF were associated with lens materials. As shown in table 5.3, Comfilcon A lens wear induced the maximum reduction of CRF followed by Stenofilcon A and Nelfilcon A. There was no significant reduction by Balafilcon A, Nesofilcon A and Lotrafilcon B lens wear. Although statistically non-significant, Comfilcon A was the only lens that steepened the anterior corneal surface. It steepened corneal surface by 0.03mm which may be clinically significant. It is not clear about the role of change in corneal shape on corneal biomechanical properties. Comfilcon A is a silicone hydrogel lens with the highest oxygen permeability and with flattest base curve among the studied lenses. It has been prepared without any surface treatment and internal wetting agent. As suggested by Szczotka-Flynn,[26] generally in silicone lenses, the higher the modulus, the higher the Dk and the higher the Dk, the lower the water content; but Comfilcon A material does not follow these trends. So, the effect of Comfilcon A wear on corneal biomechanical properties might be due to its different material chemistry.

A potential limitation of the current study could be that we did not evaluate the central corneal thickness of the eyes. However, due to daily wear modality of CL and each had oxygen transmissibility enough to avoid corneal edema,[24] any corneal thickness changes were not expected. To support this, we found no change in CH and decrease in CRF after CL wear. Previous studies suggested that corneal thickness and CRF correlate positively.[23,21] So, if there was an increase in corneal thickness, there should be an increase in CRF and this was not the case.

From the findings of this study, it can be concluded that soft CL wear reduced CRF. This effect was associated with lens materials: Comfilcon A induced greatest change among the studied materials. Earlier it was found that change in corneal biomechanical properties is reversible.[17] A long time longitudinal study is recommended to confirm the findings of this study and to investigate the permanency of the changes in CRF.

References

1. Kaushik S, Pandav SS, Banger A, Aggarwal K, Gupta A. Relationship between corneal biomechanical properties, central corneal thickness, and intraocular pressure across the spectrum of glaucoma. *Am. J. Ophthalmol.* 2012;153(5):840-849.e2.

2. Carbonaro F, Hysi PG, Fahy SJ, Nag A, Hammond CJ. Optic disc planimetry, corneal hysteresis, central corneal thickness, and intraocular pressure as risk factors for glaucoma. *Am. J. Ophthalmol.* 2014;157(2):441-446.

3. Medeiros F, Meira-Freitas D, Lisboa R, Kuang TM, Zangwill LM, Weinreb RN. Corneal hysteresis as a risk factor for glaucoma progression: A prospective longitudinal study. *Ophthalmology* 2013;120(8):1533-1540.

4. Ahmadi Hosseini SM, Abolbashari F, Niyazmand H, Sedaghat MR. Efficacy of corneal tomography parameters and biomechanical characteristic in keratoconus detection. *Contact Lens Anterior Eye* 2014;37(1):26-30.

5. Touboul D, Bénard A, Mahmoud AM, Gallois A, Colin J, Roberts CJ. Early biomechanical keratoconus pattern measured with an ocular response analyzer: Curve analysis. *J. Cataract Refract. Surg.* 2011;37(12):2144-2150.

6. González-Méijome JM, Villa-Collar C, Queirós A, Jorge J, Parafita M. Pilot study on the influence of corneal biomechanical properties over the short term in response to corneal refractive therapy for myopia. *Cornea* 2008;27(4):421-6.

7. Luce D. Determining in vivo biomechanical properties of the cornea with an ocular response analyzer. *J. Cataract Refract. Surg.* 2005;31(1):156-62.

8. Sapkota K, Franco S, Lira M. Intraocular pressure measurement with ocular response analyzer over soft contact lens. *Contact lens anterior eye* 2014;In press.

9. Shah S, Laiquzzaman M, Yeung I, Pan X, Roberts C. The use of the Ocular Response Analyser to determine corneal hysteresis in eyes before and after excimer laser refractive surgery. *Contact Lens Anterior Eye* 2009;32(3):123-128.

10. Touboul D, Roberts C, Kérautret J, *et al*. Correlations between corneal hysteresis, intraocular pressure, and corneal central pachymetry. *J. Cataract Refract. Surg.* 2008;34(4):616-22.

11. Ogbuehi KC, Osuagwu UL. Corneal biomechanical properties: Precision and influence on tonometry. *Contact Lens Anterior Eye* 2013..

12. Shah S, Laiquzzaman M. Comparison of corneal biomechanics in pre and post-refractive surgery and keratoconic eyes by Ocular Response Analyser. *Contact Lens Anterior Eye* 2009;32(3):129-132.

13. Fontes BM, Ambrósio R, Jardim D, Velarde GC, Nosé W. Corneal biomechanical metrics and anterior segment parameters in mild keratoconus. *Ophthalmology* 2010;117(4):673-9.

14. Chen MC, Lee N, Bourla N, Hamilton DR. Corneal biomechanical measurements before and after laser in situ keratomileusis. *J. Cataract Refract. Surg.* 2008;34(11):1886-91.

15. Beshtawi IM, O'Donnell C, Radhakrishnan H. Biomechanical properties of corneal tissue after ultraviolet-A-riboflavin crosslinking. *J. Cataract Refract. Surg.* 2013;39(3):451-462.

16. Chen D, Lam AKC, Cho P. A pilot study on the corneal biomechanical changes in short-term orthokeratology. *Ophthalmic Physiol. Opt.* 2009;29(4):464-471.

17. Nieto-Bona A, González-Mesa A, Villa-Collar C, Lorente-Velázquez A. Biomechanical properties in corneal refractive therapy during adaptation period and after treatment interruption: A pilot study. *J. Optom.* 2012;5(4):164-170.

18. Nathon E. Efron Grading Scales for Contact Lens Complications. In: Efron N, Ed. Contact Lens Complications. Oxford: Butterworth- Heinnemann; 2004:239-43.

19. Shen M, Wang J, Qu J, *et al*. Diurnal variation of ocular hysteresis, corneal thickness, and intraocular pressure. *Optom. Vis. Sci.* 2008;85(12):1185-92.

20. González-Méijome JM, Queirós A, Jorge J, Díaz-Rey A, Parafita M. Intraoffice variability of corneal biomechanical parameters and intraocular pressure (IOP). *Optom. Vis. Sci.* 2008;85(6):457-462.

21. Franco S, Lira M. Biomechanical properties of the cornea measured by the Ocular Response Analyzer and their association with intraocular pressure and the central corneal curvature. *Clin. Exp. Optom.* 2009;92(6):469-475.

22. Narayanaswamy A, Chung RS, Wu RY, *et al*. Determinants of corneal biomechanical properties in an adult Chinese population. *Ophthalmology* 2011;118(7):1253-1259.

23. Lu F, Xu S, Qu J, *et al*. Central corneal thickness and corneal hysteresis during corneal swelling induced by contact lens wear with eye closure. *Am. J. Ophthalmol.* 2007;143(4):616-22.

24. Holden B, Mertz GW. Critical oxygen levels to avoid corneal edema for daily and extended wear contact lenses. *Invest. Ophthalmol. Vis. Sci.* 1984;25(10):1161-7.

25. Goldich Y, Barkana Y, Pras E, *et al*. Variations in corneal biomechanical parameters and central corneal thickness during the menstrual cycle. *J. Cataract Refract. Surg.* 2011;37(8):1507-1511.

26. Szczotka-Flynn L. Lens Distinctions. *Contact Lens Spectr.* 2007;22:6.

CHAPTER 6. **A**

INTRAOCULAR PRESSURE MEASUREMENT WITH OCULAR RESPONSE ANALYZER OVER SOFT CONTACT LENS

Highlights

- **Goldmann-correlated intraocular pressure (IOPg) and corneal-compensated intraocular pressure (IOPcc) were measured with and without minus soft contact lenses (SCL) with ocular response analyzer (ORA).**
- **ORA underestimated IOPg and IOPcc when measured over SCL.**
- **Underestimation was lower with thinner and low modulus lenses.**
- **IOPg was less affected in comparison to IOPcc.**

This chapter is based on the following original article: Sapkota K, Franco S, Lira M. Intraocular pressure measurement with ocular response analyzer over soft contact lens. Contact Lens Anterior Eye, 2014;37(6):415-9.

6A.1 Introduction

There are estimated 140 million people wearing contact lens (CL) in the world for refractive purposes.[1] Many other people are also wearing CL for therapeutic purpose, since some CLs were found to be effective in pain relief, corneal healing or mechanical support in some corneal diseases: persistent epithelial defects, recurrent corneal erosions, filamentous keratitis, corneal thinning, bullous keratopathy.[2,3]

For the complete ocular examination and the follow-up examinations of glaucoma susceptible patients, accurate intraocular pressure (IOP) measurement is important. Measurement of IOP by Goldmann Applanation Tonometry (GAT) is the gold standard and is being used since many years with good accuracy.[4] However, IOP measured by GAT is dependent upon the corneal biomechanical properties.[5,6] Moreover, in many countries, optometrists are not allowed to use anesthetic drop and fluorescein dye which are necessary for GAT. So, non-contact tonometry is popular for many practitioners.

The Ocular Response Analyzer (ORA; Reichert Ophthalmic Instruments, Buffalo, NY) measures IOP, regardless of corneal biomechanical properties.[6] The principle of the ORA is based on those of non-contact tonometry, in which the IOP is determined by the air pressure required to applanate the central cornea. The detailed information about the ORA can be found in other studies.[7] Briefly, this instrument utilizes a rapid air impulse to deform the cornea during which the shape of the cornea is monitored by an electro-optical system. The instrument fires a metered collimated air pulse at the cornea so that the convex-shaped cornea changes to plane (inward applanation) and to slight concave shape. After the air puff pressure reduces, the cornea comes again to plane shape (outward applanation) and to the convex shape as normal. All these processes complete within 20-25 milliseconds. The inward applanation pressure is always less than the outward applanation pressure due to some energy absorption by cornea.[7] The average of these two pressures is Goldmann-correlated intraocular pressure (IOPg) which is correlated with pressure measured by Goldmann applanation tonometry.[8] The corneal-compensated intraocular pressure (IOPcc) is calculated with linear relationship of these two pressures and is considered to be less affected by corneal thickness and corneal biomechanical properties.[6]

Patients, who wear CL, usually remove them before IOP measurements. Removal of CL may cause temporal changes in IOP and may cause impairment in epithelization process in some

cases.[9] Many people may not desire to remove the CL for IOP measurements, especially if it is required frequently. In many countries, optometrists are not allowed to use anesthetic eye drop. So, if measurement of IOP over CL is accurate, it can be considered as an option in these situations.

Many studies have been done to evaluate the IOP with and without CL using different methods of tonometry as summarized in table 6A.1.[10-21] Some of them found significant differences in the two measurements,[11,13,14,17,21] while others did not find any differences.[10,12,15,16,20]

However, in our knowledge, no studies have been conducted comparing IOP with and without CL using ORA. One of the aims of this study was to investigate the influence of soft CL in the IOP measurement by ORA. Another aim of this study was to determine -- out of IOPg and IOPcc -- which one is less affected by the presence of CL. If there is no clinically significant difference, there is no need to remove soft CL before IOP measurements.

6A.2 Methods

A cross-sectional prospective study was conducted in normal subjects recruited from University of Minho, Portugal. A general primary ocular examination was done and, subjects having any ocular pathology, ocular surgery and those apparently normal but resulting in IOPg or IOPcc values more than 21 mmHg in at least one eye, were excluded.

This study was approved by the School of Science Ethical Committee, University of Minho. All the subjects gave informed consent after nature of the study had been explained and the tenets of the Declaration of Helsinki were followed.

Table 6A.1. Preview of studies comparing the intraocular pressure measured with and without contact lens with different tonometry

Author (year)	N (eyes)	Measurement technique	Material (parameter)	Remarks
Gogniat *et al*.[10] (2013)	42	DCT	Nelfilcon A, Narafilcon A (P = +5.00D, - 0.50D and - 5.00D)	No significant difference except for +5.00D Nelfilcon A
Schollmayer *et al*.[11] (2003)	120	Non-contact pneumotonometry	Lotrafilcon A (P = -1.00D, -4.00D, +1.00D, +4.00D)	Underestimation in minus lenses Overestimation in plus lenses The difference was correlated with the power of the lenses
Zeri *et al*.[12] (2007)	136	GAT	Hilafilcon A (parameter not available)	No significant difference
Patel *et al*.[13] (2009)	50	NCT	Lotrafilcon A and Nelfilcon A	Minus lenses: underestimation plus lenses: overestimation Difference correlated with power
Liu *et al*.[14] (2011)	32	NCT	Hilafilcon A (P = -3.00D to -12.00D)	Underestimation, correlated with power
Allen *et al*.[15] (2007)	20	GAT	Silicone hydrogel lenses (parameter not available)	No significant difference
Schornack *et al*.[16] (2012)	78	Tonopen XL	Galyfilcon A, Senofilcon A, Lotrafilcon B (P = -0.25D to -3.00D and -3.25D to -6.00D)	No significant difference except with high power Lotrafilcon B lenses
Boyraz *et al*.[17] (2013)	30	Tonopen XL	Lotrafilcon A, Balafilcon A and Vifilcon A (P = -3.00D)	Overestimation
Anton *et al*.[18] (2013)	39	ICare rebound tonometry and Airpuff tonometry	Therapeutic soft lenses (materials not available)	ICare: overestimation Airpuff: no difference

[GAT – Goldmann applanation tonometry, NCT – non-contact tonometry, DCT – dynamic contour tonometry, P - Power]

Initially, IOPg and IOPcc were measured in both the eyes with ORA. Tonometry was done initially without CL on the eye to prevent the possible alteration in intraocular pressure due to change in corneal curvature immediately after CL removal.[22] After that, subjects were fitted with a silicone hydrogel lens (Narafilcon A) in one eye (Group A) and a hydrogel lens (Nelfilcon A) in the other eye (Group B) each with -3.00D. These lenses were chosen because of their different material properties and designs (CL details are specified in table 6A.2). After 10 minutes of CL wear, as in a previous study,[13] IOPg and IOPcc were measured over the CL by the same investigator using the same instrument. For all the measurements, three readings were taken and average was used in the subsequent analysis. All these assessments were done at 14:00-17:00 hours.

Data were analyzed with IBM SPSS 21 statistical software (IBM Corp., Armonk, NY). Kolmogorov-Smirnov test was used to evaluate the normality of the data distribution. Parametric tests were applied for the normally distributed variables and non-parametric tests for the others. Pearson correlation test was applied to determine the correlation in IOPg and IOPcc measurements with and without CL. Paired sample test was applied to determine the variation of IOP with and without CL. Bland-Altman plots were used to assess the variation in IOP without and with CL as a function of IOP value. For all the analysis, $p \leq 0.05$ was considered as statistically significant.

Table 6A.2. Details of the contact lenses used in the study.

Parameters	Group A	Group B
Company name	Johnson & Johnson	Ciba Vision
Brand name	1-Day Acuvue® True Eye™	Daily® AquaComfort Plus®
Material	Narafilcon A	Nelfilcon A
Power (D)	-3.00	-3.00
Water content	46%	69%
Base curve/Diameter (mm)	8.5/14.2	8.7/14
Oxygen permeability (Barrer)	100	26
Center thickness (mm)	0.085	0.10
Modulus (MPa)	0.66	0.89

6A.3 Results

A total of 28 subjects with mean (±standard deviation) age of 29.4 ± 9.8 years have participated in this study. Fifty-four percent (15) were female. None of the subjects were wearing CL or spectacle before.

Both IOPg and IOPcc presented lower values for the measurements done over the lens in both types of CL than that of the measurements without CL (Table 6A.3).

These differences were higher for the Nelfilcon A CL in comparison to Narafilcon A but not statistically significant ($p > 0.05$). The values of IOPcc were more affected than IOPg although the differences were not statistically significant ($p>0.05$) [Figure 6A.1].

Table 6-3. Intraocular pressures without and with contact lens

		Intraocular pressure Mean ± standard deviation [Range]			**P value**
		Without contact lens	**With contact lens**	**Difference**	
Narafilcon A	**IOPg (mmHg)**	14.13±2.85 [9.23-18.57]	13.24±3.74 [6.07-20.87]	0.85±2.00 [-4.17-5.33]	0.026
	IOPcc (mmHg)	4.15±3.18 [8.67-20.57]	12.60±3.66 [6.10-20.97]	1.55±2.16 [-3.07-4.87]	0.001
Nelfilcon A	**IOPg (mmHg)**	14.10±2.62 [10.13-18.27]	13.06 ±2.93 [7.50-18.63]	1.03±1.93 [-2.87-5.33]	0.009
	IOPcc (mmHg)	13.93±3.19 [7.10-19.27]	12.31±3.48 [4.07-19.63]	1.62±3.12 [-4.53-10.03]	0.011

IOPg- Goldmann–correlated intraocular pressure, IOPcc- corneal–compensated intraocular pressure.

Both the IOPg and IOPcc with and without CL were highly positively correlated in both Narafilcon A and Nelfilcon A lenses ($p < 0.001$).

The difference was within 3mmHg in 82% of the IOPg measurements with Narafilcon A; 86% of the IOPg values with Nelfilcon A; 64% of the IOPcc values with Narafilcon A; 79% of the IOPcc values with Nelfilcon A.

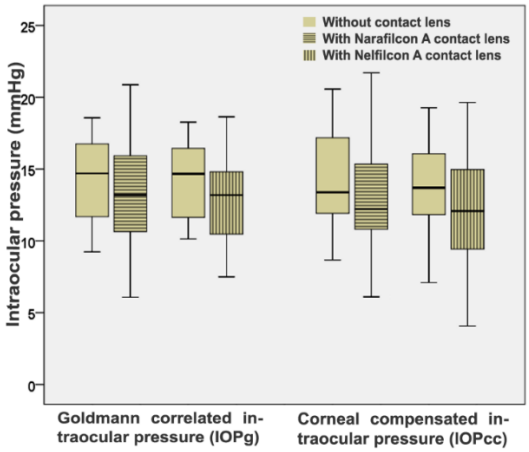


Figure 6A.1. Intraocular pressure without and with contact lenses

Graphical analysis of difference against mean as proposed by Bland and Altman (1986) is shown in figures 6A.2 and 6A.3. In these graphics, the difference between the two IOP (IOP without CL – IOP with CL) is plotted against its mean, for each type of CL. This analysis was done for both IOPg (Figure 6A.2) and IOP_{CC} (Figure 6A.3).

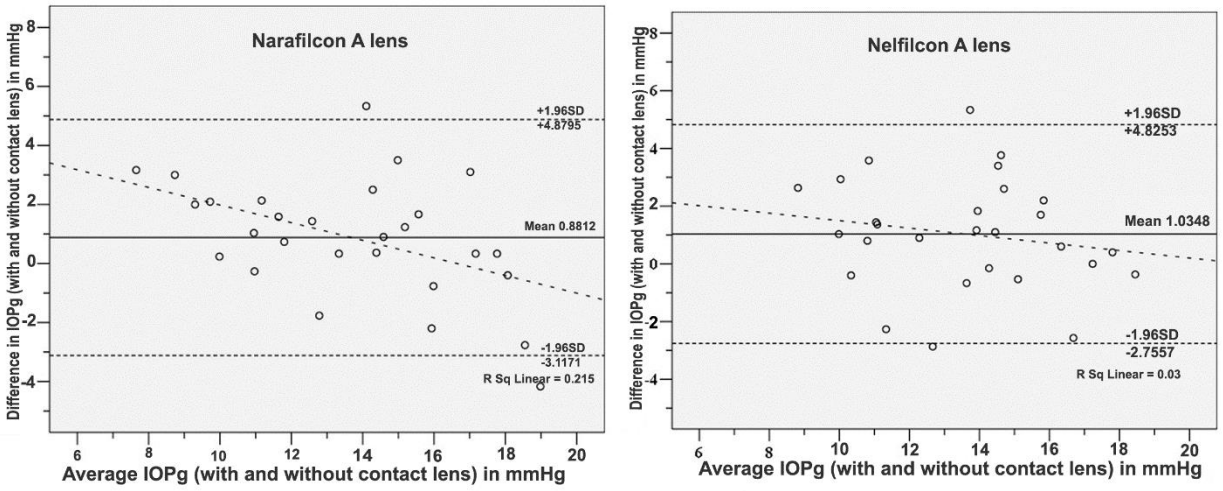


Figure 6A.2. Bland-Altman Plot of IOPg (with Narafilcon A and Nelfilcon A lens). Dash horizontal lines showing limit of 95% confidence interval [IOPg- Goldmann-correlated intraocular pressure].

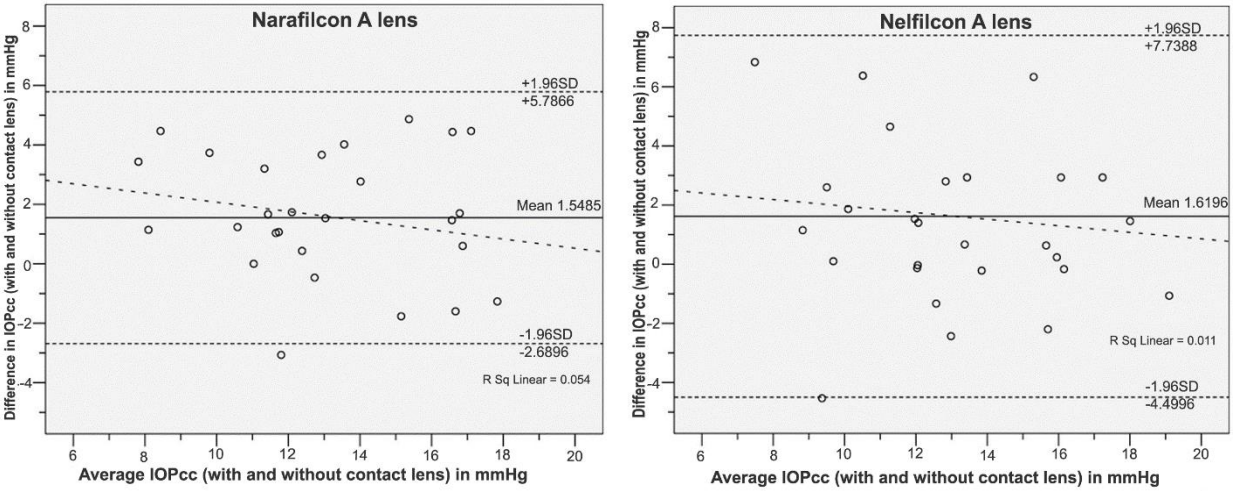


Figure 6A.3. . Bland-Altman Plot of IOPcc (with Narafilcon A, and Nelfilcon A lens). Dash horizontal lines showing limit of 95% confidence interval [IOPcc – corneal-compensated intraocular pressure].

With this type of plot, it is easier to assess the magnitude of disagreement and see whether there is any tendency for the difference between the two methods. For each CL type, it is also represented the mean difference between the measurements obtained with and without CL (solid line) as well as the limits of agreement (dashed lines) which are given by the mean ± 1.96x SD.

For Narafilcon A, figure 6.1.2 shows a mean difference of 0.88 mmHg for the IOP_g between values measured with and without the CL, with a significant underestimation of the values obtained over the CL, with the limits of agreement varying from +4.9 mm Hg to -3.11 mmHg. The Nelfilcon A CL presents a similar behavior. However, the underestimation is higher (mean of 1.03 mmHg) and the limits of agreement smaller (+4.82 mmHg to -2.76 mmHg).

The IOPcc analysis shows a similar behavior. For both CL materials, the difference in the IOP_{cc} values with and without CL was higher. Additionally, from all the plots it is also possible to see that no significant tendency has been identified for differences between the measures of both IOP with and without CLs as a function of their mean value. Only for the IOPg for the Narafilcon A lens presents a negative tendency ($r = - 0.469$, $p = 0.012$). As the mean values increase the differences between the two measurements decrease, being negative for mean values superior to 14 mmHg. This means that there is a tendency for an overestimation of the values with the CL for IOPg values over 14 mmHg.

6A.4 Discussion

We compared both IOPg and IOPcc using ORA in eyes with and without CL of different materials and designs with the aim of determining if there are differences in the IOP measurements. IOPg and IOPcc were found statistically significantly underestimated in both groups when measured over CL. This shows that ORA (which is considered as independent on the corneal properties) depends on the contact lens material properties on measuring IOP. IOPcc in both the types of CL is highly underestimated by the presence of the CL. As suggested by Liu *et al*., the underestimation of the IOP may be due to the reduction in the time needed to achieve the maximal light detection in NCT when the front surface is flattened by an air puff.[14]

Our findings support the results of many other studies [Table 6.1.1] which also found the underestimation of IOP over soft CL.[11,14] Liu *et al* found that minus CL underestimates the IOP measured by non-contact tonometry.[14] Schollmayer and Hawlina measured IOP with and without CL in 120 eyes by non-contact pneumotonometry.[11] They also found that minus CL

underestimates the IOP. Firat *et al* also found significant under-estimation of IOP over silicone hydrogel CL.[21]

However, some of the studies showed no significant difference in the IOP measurement with and without CL.[10,12,20] Gogniat *et al* investigated the reliability of IOP measured by Dynamic contour tonometry with and without contact lens.[10] They also used the same material as our study – hydrogel lens with Nelfilcon A and silicone hydrogel lens with Narafilcon A with varying power: +5.00D, -0.50D and -5.00D. They did not find any difference between the IOPs with and without contact lens. Zeri *et al* conducted a study in which IOP measured by GAT with and without low modulus minus power daily disposable CL were compared.[12] They did not find any significant difference.[12] In our study also, IOPg was found less affected than that of IOPcc.

As such, according to our results, and previous studies, non-contact tonometer under-estimates IOP over minus CL while Tonopen, iCare Rebound Tonometer over-estimate IOP over CL.[11,13,14,17,18]

Patel and Stevenson found that the variation in the IOP measurements using a non-contact tonometer with and without CL depends on the lens materials.[13] In our study, we did not get any significant difference between the two types of materials. However, the variation was slightly higher with Nelfilcon A than with Narafilcon A lens. This may be due to the facts that, Nelfilcon A lenses have greater center thickness and higher modulus. Zeri *et al* also found higher difference in hydrogel lenses than that of silicone lenses.[20]

McMonnies found that IOP measurement does not significantly differ with soft CL with center thickness less than 0.15 mm.[19] But, statistically, significant difference was found in our study when IOP was measured with and without CL, though central thickness was less than 0.15 mm. However, only hydrogel lenses were used in that study while we had used both hydrogel and silicone hydrogel lenses.

Bland Altman plots showed that almost all the data points were within 2 standard deviations from the mean difference between IOP without and with CL. No significant trend was found for the variations in two IOP measures as a function of IOP value. The mean difference was 0.85 mmHg to 1.62 mmHg which can be considered clinically insignificant as

the variation in IOP is clinically acceptable in a range of ±3 mmHg.[23],[24] However, IOPcc was underestimated by more than 3 mmHg in up to 36% of the patients.

There are some limitations in this study. We included only those eyes having IOP in normal range. So the findings of this study may not be equally applied for the eyes with abnormal IOP.

In summary, the reliability of IOP measured over soft contact lens has given conflicting results in the literature [Table 6A.1]. This might be because of the variation in types of tonometry (instruments), lens parameters (central thickness and power) and the lens materials. So it can be concluded that the variation is due to the type of tonometry besides the lens materials and parameters. Our study shows that ORA over minus (-3.00D) CL underestimates the IOP and the underestimation is higher in CL with higher thickness and modulus. In clinical practice, CL should be removed before tonometry for the accurate IOP measurement. If tonometry is done over CL by ORA, IOPg should be considered, seeing that it is less affected by CL.

References

1. Stapleton F, Keay L, Jalbert I, Cole N. The epidemiology of contact lens related infiltrates. *Optom. Vis. Sci.* 2007;84(4):257-72.

2. Coral-Ghanem C, Ghanem VC, Ghanem RC. Therapeutic contact lenses and the advantages of high Dk materials. *Arq. Bras. Oftalmol.* 2008;71:19-22.

3. Rubinstein MP. Applications of contact lens devices in the management of corneal disease. *Eye* 2003;17(8):872-876.

4. Sudesh S, Moseley MJ, Thompson JR. Accuracy of Goldmann tonometry in clinical practice. *Acta Ophthalmol.* 1993;71(2):185-8.

5. Colás-Tomás T, Prieto-Del Cura M, Villafruela-Güemes I, Clariana-Martín A, Valdivia-Pérez A. [Comparison of dynamic contour tonometry, Goldmann and pneumotonometer in ocular hypertension patients and their relationship to pachymetry and ocular pulse amplitude]. *Arch. Soc. Esp. Oftalmol.* 2012;87(12):401-6.

6. Neuburger M, Maier P, Böhringer D, Reinhard T, F Jordan J. The impact of corneal edema on intraocular pressure measurements using goldmann applanation tonometry, Tono-Pen XL, iCare, and ORA: an in vitro model. *J. Glaucoma* 2013;22(7):584-90.

7. Oliveira C, Franco S. Measuring Cornea. In: *Advance in Eye Research*.; 2012:115-36.

8. Fabian ID, Barequet IS, Skaat A, *et al*. Intraocular pressure measurements and biomechanical properties of the cornea in eyes after penetrating keratoplasty. *Am. J. Ophthalmol.* 2011;151(5):774-81.

9. Khan JA, Graham CE. Effect of contact lens removal or displacement on intraocular pressure. *Arch. Ophthalmol.* 1991;109(6):825-828.

10. Gogniat F, Steinegger D, Nosch D, Joos R, Goldschmidt M. The Accuracy of Dynamic Contour Tonometry. *Optom. Vis. Sci.* 2013;90(2):125-130.

11. Schollmayer P, Hawlina M. Effect of Soft Contact Lenses on the Measurements of Intraocular Pressure with Non-Contact Pneumotonometry. *Klin. Monbl. Augenheilkd.* 2003;220(12):840-842.

12. Zeri F, Lupelli L, Formichella P, Masci C, Fletcher R. Goldmann applanation tonometry over daily disposable contact lens: Accuracy and safety of procedure. *Contact Lens Anterior Eye* 2007;30:233–238.

13. Patel S, Stevenson G. Influence of lens material and intra-ocular pressure on the outcome of non-contact tonometry over soft contact lenses. *Contact Lens Anterior Eye* 2009;32(2):68-72.

14. Liu Y-C, Huang J-Y, Wang I-J, Hu F-R, Hou Y-C. Intraocular pressure measurement with the noncontact tonometer through soft contact lenses. *J. Glaucoma* 2011;20(3):179-182.

15. Allen RJ, Dev Borman A, Saleh GM. Applanation tonometry in silicone hydrogel contact lens wearers. *Contact Lens Anterior Eye* 2007;30(5):267-269.

16. Schornack M, Rice M, Hodge D. Tonopen XL Assessment of Intraocular Pressure Through Silicone Hydrogel Contact Lenses. *Eye Contact Lens Sci. Clin. Pract.* 2012;38(5):270-273.

17. Boyraz S, Güngör I. The effects of the modulus of the lens material on intraocular pressure measurement through soft contact lenses. *Ir. J. Med. Sci.* 2013;182(3):331-335.

18. Anton A, Neuburger M, Böhringer D, Jordan JF. Comparative measurement of intraocular pressure by Icare tonometry and Airpuff tonometry in healthy subjects and patients wearing therapeutic soft contact lenses. *Graefe's Arch. Clin. Exp. Ophthalmol.* 2013;251(7):1791-1795.

19. McMonnies CW. Noncontact tonometry through soft contact lenses. *Am. J. Optom. Physiol. Opt.* 1986;63(12):948-951.

20. Zeri F, Calcatelli P, Donini B, Lupelli L, Zarrilli L, Swann PG. The effect of hydrogel and silicone hydrogel contact lenses on the measurement of intraocular pressure with rebound tonometry. *Contact Lens Anterior Eye* 2011;34(6):260-265.

21. Fırat PG, Cankaya C, Doganay S, *et al*. The influence of soft contact lenses on the intraocular pressure measurement. *Eye* 2012;26(2):278-282.

22. Jorge J, González-Méijome JM, Queirós A, Fernandes P, Diaz-Rey JA. A comparison of the NCT Reichert R7 with Goldmann applanation tonometry and the Reichert ocular response analyzer. *Ophthalmic Physiol. Opt.* 2011;31(2):174-9.

23. Christoffersen T, Holtedahl K, Ringberg U, Fors T. Can the Tono-Pen replace the Schiøtz tonometer in general practice? *Scand. J. Prim. Health Care* 1998;16(4):238-241.

24. Lam AKC, Lam CH, Chan R. The validity of a digital eyelid tonometer (TGDc-01) and its comparison with Goldmann applanation tonometry - A pilot study. *Ophthalmic Physiol. Opt.* 2005;25(3):205-210.

CHAPTER 6. **B**

EFFECT OF THREE MONTHS OF SOFT CONTACT LENS WEAR ON INTRAOCULAR PRESSURE

Highlights

- **Effects of soft contact lens (SCL) wear on intraocular pressure (IOP) was investigated.**
- **IOP reduced significantly after three months of SCL wear.**
- **It was higher during earlier period.**
- **IOP reduction was similar between daily disposable and monthly disposable lens wear but was different with different lens materials.**
- **Higher reduction was observed in corneas with higher corneal resistance factor.**

This chapter is based on the following original article: Sapkota K, Franco S, Lira M. Effect of three months of soft contact lens wear on intraocular pressure. Submitted to Contact Lens Anterior Eye in September 2015.

6B.1 Introduction

The number of contact lenses (CL) wearers is exceeding 140 million in the world.[1] Besides the use of CL for refractive error correction, many people wear CL for therapeutic[2] and aesthetic purposes.[3] Recent studies found positive results of use of CL on continuous drug delivery into the eyes[4] and on monitoring tear glucose level.[5]

CL wear can have an effect upon the cornea, as lenses are directly placed over it. It may cause corneal swelling as a consequence of corneal hypoxia.[6] This effect is prominent in cases of extended wear with low gas permeable CL.[7] However, silicone-hydrogel lenses, regardless of wearing modality, reduce the hypoxia-related problems.[8] Due to inhibition of corneal epithelial mitosis and the physical weight and tension of lids over CL,[9] it may lead to corneal thinning.[10] Studies show that corneal thickness affects intraocular pressure (IOP): a thicker cornea overestimates the IOP and a thinner cornea underestimates it.[11,12] Some researchers[13] found that CL wear changes corneal biomechanical properties like corneal hysteresis (CH) and corneal resistance factors (CRF) which are supposed to have an important role on measurement of IOP.[14] Corneal curvature may also affect IOP measurement.[15] Thus, CL wear can influence IOP due to the changes in corneal characteristics.

Although Goldmann applanation tonometry (GAT) is considered as the gold standard for IOP measurement and as having good accuracy,[16] recent studies found that IOP measured by GAT is dependent on corneal biomechanical properties.[17,18] Moreover, in many countries, optometrists are not allowed to use an anesthetic drop and fluorescein dye which are necessary in GAT. As such, non-contact tonometry is popular for many practitioners. Ocular Response Analyzer (ORA; Reichert Ophthalmic Instruments, Buffalo, NY) measures IOP regardless of corneal biomechanical properties.[18] It provides readings of Goldmann-correlated IOP (IOPg) and corneal-compensated IOP (IOPcc) as well as biomechanical properties of the cornea: corneal hysteresis (CH) and corneal resistance factor (CRF). The principle of ORA is based on those of non-contact tonometry, in which IOP is determined by the air pressure required to applanate the central cornea. Detailed information about ORA can be found elsewhere.[19] Briefly, this instrument uses a rapid air impulse to deform the cornea during which the shape of the cornea is monitored by an electro-optical system. The instrument fires a metered

collimated air pulse at the cornea so that the convex-shaped cornea changes to plane (inward applanation) and to slight concave shape. After the air puff pressure reduces, the cornea comes again to plane shape (outward applanation) and to convex shape as normal. The inward applanation pressure is always less than the outward applanation pressure due to some energy absorption by the cornea.[20] The average of these two applanation pressures is IOPg which is correlated with the pressure measured by GAT.[21] IOPcc which is considered to be less affected by corneal thickness and corneal biomechanical properties, is the linear relationship of these two pressures.[18]

Mahjoob *et al*.[22] found a significant decrease in IOP with soft and hard CL wear. Similarly, Oh *et al*.[23] found a significant decrease in IOP with soft CL wear. In these studies, CL were worn only for two hours for research purpose to induce corneal edema and GAT was used to measure IOP. To the authors' knowledge, no studies have been done in the past to investigate the effect of CL, worn for the refractive purpose, on the IOP. The aim of this study was to determine the effect of three months of soft CL wear on IOPg and IOPcc. We also studied the effect of materials and wearing modality of the CL on IOP. This study sought to find out the role of corneal biomechanical properties (CH, CRF) as well as corneal curvature on the change of IOP.

6B.2 Methods

This was a contra-lateral longitudinal clinical trial conducted in University of Minho, Portugal. It was approved by Ethical Committee of the University of Minho. Before the study began, details about the study protocol were explained and a consent form was signed by each subject. Tenets of Declaration of Helsinki were followed.

Subjects over the age 18, myopic refractive error with astigmatism less than 1.00D and best corrected visual acuity of 20/20 or better were included in this study. Previous CL wearers and subjects with ocular pathology and systemic illness, as well as those with past history of ocular surgery were excluded.

IOPg and IOPcc were measured in each eye of every subject with ORA. CH and CRF were also recorded during the baseline visit. The examination was performed with the patient

seated in a comfortable position. Three measurements were taken in each eye, and the average of these measurements was used for the analysis. To ensure good quality of measurements, only readings with well-defined applanation peaks, fairly symmetrical and regular in height were considered. Irreproducible out of scale measurements, asymmetric or wider or irregular peaks indicating an abnormal corneal movement or surface irregularity during the examination were discarded. Corneal curvature was measured by Medmont E-300 (Medmont Pty., Ltd., Melbourne, Australia). Subjects were fitted with a daily disposable lens (Nelfilcon A or Stenofilcon A or Nesofilcon A) in one eye and a monthly disposable lens (Lotrafilcon B or Comfilcon A or Balafilcon A) on daily wear modality in the other eye. Lens details are presented in table 6.2.1. Post-lens fitting evaluation was performed and refitting with another type of CL among the study lenses was done where the fitting was unacceptable. Subjects were well trained for CL usage, care and maintenance. CL, lens case and multipurpose solution [OPTI-FREE Puremoist (Polyquad 0.001% and Aldox 0.0006%, Alcon Laboratories, TX) or AO-SEPT PLUS (Hydrogen peroxide 3%, Alcon Laboratories, TX)] were provided for each subject for the coming month. Moreover, a sheet of paper containing detail information about the lens for the right eye and left eye was provided to each subject in which they should note the number of wearing hours every day. They were suggested to wear lenses 8 hours per day and 5 days per week in minimum. During monthly follow-up visits, compliance of the subjects to the study protocol was monitored. For the first month, all the subjects used OPTI-FREE Puremoist solution while for the second and third months, each subject used either OPTIFREE Puremoist or AOSEPT PLUS which was randomly selected. These two types of solutions were provided for each subject since another objective of the study was to investigate the effect of different types of solutions in CL wearers' comfort (to be published). Subjects were advised to contact the researcher at any time if they felt adverse events for the necessary management. IOPg and IOPcc were measured on each monthly follow-up visit. Measurements were taken after CL removal and on the same time of the day during all the follow-up visits.

Data were analyzed with Statistical Package for Social Science (SPSS 22, IBM Corp., Armonk, NY). Descriptive data were expressed in mean ± standard deviation (SD). Normality of the data was tested by Kolmogorov-Smirnov test. Parametric tests were applied for normally distributed variables and non-parametric tests for other variables. Repeated measure of ANOVA was performed to test the changes in IOP before and after CL wear. IOP was

compared with different lens materials (daily versus monthly and hydrogel versus silicone hydrogel) by one-way ANOVA. Bland-Altman Plotting was done to compare the IOP before and after CL wear. P values less than 0.05 were considered as statistically significant. The changes in IOP in right and left eyes were not correlated ($p > 0.05$), so data of both the eyes in each subject were used in the analysis.Table 6B.6-1. Characteristics of the contact lenses used in the study

Table 6B.1. Characteristics of the contact lenses used in the study

	AirOptix ® Aqua™	**Dailies® AquaComfort Plus®**	**Biofinity™**	**MyDay™**	**Purevision2™**	**Biotrue™ ONEday**
Company	Alcon	Alcon	Cooper Vision	Cooper Vision	Bausch & Lomb	Bausch & Lomb
Material	Lotrafilcon B	Nelfilcon A	Comfilcon A	Stenofilcon A	Balafilcon A	Nesofilcon A
Water content (%)	34	69	48	54	36	78
Thickness (mm)	0.08	0.10	0.08	0.08	0.07	0.1
Base curve/ diameter (mm)	8.6/14.2	8.7/14	8.7/14.5	8.4/14.2	8.6/14	8.6/14.2
Oxygen Permeability (barrer)	110	26	128	80	91	42
Modulus (MPa)	1.2	0.89	0.75	0.4	1.1-1.25	0.49
Transmisibility (barrer/cm)	137.5	26	160	100	130	42

6B.3 Results

Ninety-four eyes of 47 subjects (66.0% female) were included. Mean age of the subjects was 24.3±4.1 years (ranges 19-35 years). Neophyte CL wearers wore a daily disposable lens in one eye and a monthly disposable lens in the other eye in contra-lateral manner with a random total distribution as follow: Nelfilcon A (n = 16), Stenofilcon A (n = 15), Nesofilcon A (n = 16), Lotrafilcon B (n = 16), Comfilcon A (n = 15) Balafilcon A (n = 16). The mean power of CL was -1.86±1.54D (range -0.50D to -4.50D). There was no significant difference in baseline values of IOPg and IOPcc within different lens group ($p > 0.05$). Subjects wore lenses for 11.83 ± 1.81 hours per day and 6.10±0.73 days per week in average.

The means of the IOPg and IOPcc were 13.49 ± 3.46 and 13.72 ± 3.06 mmHg during baseline visit while the values were 11.72 ± 2.85 and 12.12 ± 2.94 mmHg respectively on the final visit. A significant reduction was found in IOPg [paired sample T test, $p < 0.001$] and IOPcc [$p < 0.001$] after CL wear. IOPg reduced 1.8 ± 2.5 mmHg and IOPcc reduced 1.6 ± 2.7 mmHg during this three months period. Figure 6B.1 shows the trend in changing IOPg and IOPcc with soft CL wear. After the first month, IOPg reduction was 0.69 ± 2.40 mmHg ($p = 0.007$) whereas after the second month it was 0.78 ± 2.18 mmHg ($p = 0.002$). However, after the third month, the reduction in IOPg was 0.27 ± 2.16 mmHg from the values of the previous month was not statistically significant ($p = 0.262$). Similarly, IOPcc reduced by 0.58 ± 2.57 mmHg ($p = 0.035$) after the first month, 0.82 ± 2.50 mmHg ($p = 0.003$) in the second month, and 0.20 ± 2.38 mmHg ($p = 0.443$) after the third month in comparison to the values of the previous month.

Change in IOPg was correlated with CRF ($r = 0.466$. $p < 0.001$) but not correlated with CH ($r = 0.195$, $p = 0.062$). Similarly, change in IOPcc was correlated with CRF ($r = 0.241$, $p = 0.021$) but not with CH ($r = -0.039$, $p = 0.713$). IOPg reduction was not correlated with corneal curvature ($r = 0.102$, $p = 0.335$). Similarly, IOPcc decrease was not correlated with corneal curvature ($r = 0.075$, $p = 0.478$)

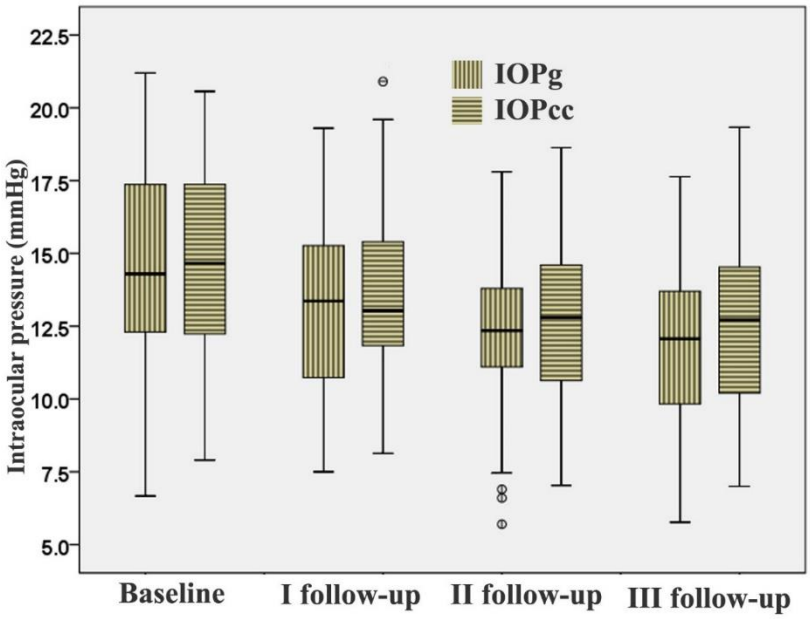


Figure 6B.1. Change in intraocular pressure with soft contact lens wear (n = 94) [IOPg: Goldmann-correlated intraocular pressure, IOPcc: corneal-compensated intraocular pressure].

Decrease in IOPg as well as IOPcc were significantly related with CL types ($p < 0.05$). Table 6B.2 shows the decrease in IOPg and IOPcc for each CL material. Comfilcon A and Stenofilcon A lenses wearers presented higher changes in IOP when compared with other lens wearers ($p < 0.05$). There was a smaller change in IOP in Balafilcon A and Nesofilcon A lens wearers which was non-significant statistically ($p > 0.05$). The changes in IOPg and IOPcc were not associated with wearing modality of CL, being similar in monthly and daily disposable lenses ($p > 0.05$). The changes were also similar with different a solution using groups ($p > 0.05$).

IOP variation (both IOPg and IOPcc) was not correlated with the age of subjects and CL power ($p > 0.05$). It was also not associated with gender ($p > 0.05$). Figure 6B.2 shows the Bland-Altman Plots of changes in IOP before and after CL wear. Almost all the points fall within the 95% confidence interval of the difference in IOPg/IOPcc. The trend shows that, on the normal range of IOP, CL wear always reduces IOP and the reduction is directly proportional to the baseline IOP.

Table 6B.6-2. Reduction in intraocular pressure after three months of different soft contact lenses wear

Lens material	Δ IOPg (mmHg)	p values	Δ IOPcc (mmHg)	p values
Lotrafilcon B	1.35±1.82	0.010	1.13±1.87	0.029
Comfilcon A	3.75±2.51	0.000	3.22±2.53	0.000
Nelfilcon A	1.68±2.27	0.010	1.35±2.54	0.050
Stenofilcon A	3.31±2.47	0.000	3.16±2.63	0.000
Balafilcon A	0.24 ± 1.98	0.646	0.14±2.82	0.847
Nesofilcon A	0.52±2.38	0.411	0.80±2.88	0.298

IOPg: Goldmann-correlated intraocular pressure, IOPcc: corneal-compensated intraocular pressure.

6B.4 Discussion

The effect of three months of soft CL wear on IOP in neophyte CL wearers was investigated and a reduction was found in IOP, regardless of CL type and wear modality. The reduction was more significant during the first two months. Because all subjects were neophyte CL wearers, the effect of CL might be higher during the initial adaptation period. In the third-month follow-up visit, the reduction was not significant when compared with IOP on the previous month. So the trend in the IOP changes with soft CL wear shows that initially it changes highly and latter slowly.

IOP decrease may be due to the effect of CL on corneal biomechanical properties. Reduction in IOPg and IOPcc was correlated with CRF. CRF shows the elastic properties of the cornea and indicates the overall resistance to the deformation.[24] A previous study found that CRF is lower in weak corneas in comparison to those of normal eyes.[25] So the findings of the current study may suggest that reduction in IOP can be lower in weaker corneas and vice-versa. Any relation in IOP reduction and anterior corneal curvature was also investigated. Changes in both IOPg and IOPcc were not correlated with corneal curvature.

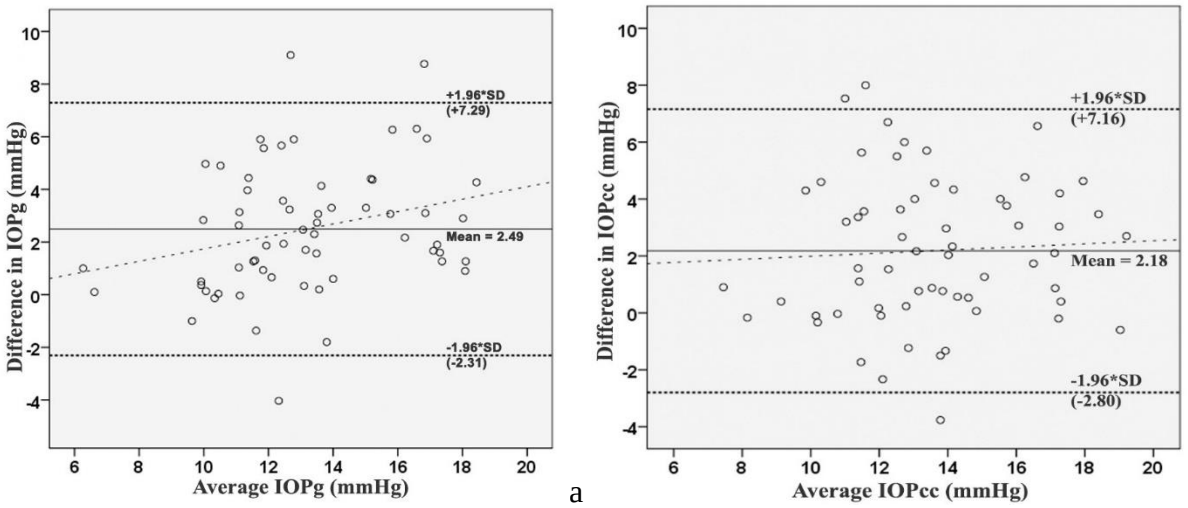


Figure 6B.2. Bland-Altman Plotting of changes in IOP before and after 3 months of contact lens wear. A. IOPg; B. IOPcc. Two parallel dotted lines show the 95% limit of confidence interval while oblique line shows the trend of changes in different amount of average IOP [IOPg: Goldmann-correlated intraocular pressure, IOPcc: corneal-compensated intraocular pressure].

Another reason of the IOP change by CL wear may be due to the alteration in corneal thickness.[10] However, it may not be the important factor behind the IOP reduction in this current study as CL were worn in daily wear modality.[26] All the lens materials had Dk/t more than 24 units, which avoids corneal swelling when lenses are worn under open-eye conditions.[27]

In the current study, no relation was observed between reduction in IOP and age of the subjects or power of the lenses. This might be due to a small range of age of the subjects and the small range of the power of the CL used in this study. Reduction in IOP was not associated with wearing modality but was associated with CL types. As can be seen from table 6.2.1 and table 6.2.2, there was not a direct relation of reduction in IOP and the lens modulus or oxygen transmissibility or water content of the lens materials. This may indicate that the cause of reduction in IOP may be multifactorial. Eyes wearing Comfilcon A lenses were found to have the highest reduction among studied eyes. Comfilcon A is a silicone hydrogel lens with the highest oxygen permeability and with flattest base curve among the studied lenses. It has been prepared without any surface treatment and internal wetting agent. As suggested by Szczotka-

Flynn,[28] generally in silicone lenses, the higher the modulus, the higher the Dk and the higher the Dk, the lower the water content; but Comfilcon A material does not follow both of these trends. So, the effect of Comfilcon A wear on IOP might be due to its different material chemistry. It can be observed that the lenses manufactured from the same companies have a similar reduction in IOP [Table 6.2.2]. This highlights that reduction in IOP is associated with the lens manufacturing process and lens design.

Bland-Altman Plots [Figure 6B.2] explain the changes in IOPg and IOPcc before and after CL wear. The trend in IOPg changes shows that the higher the baseline IOPg, the higher the reduction by CL wear; however, initial IOPcc has little effect on the change in IOPcc. The plots also highlight that soft CL wear reduces IOPg and IOPcc in the normal range of IOPg/IOPcc and the changes were found within 95% confidence interval of the standard deviation of the mean reduction.

The potential limitation of this clinical trial could be that we did not measure corneal thickness. However, since all CL were worn in daily modality; four CL used in this study were silicone hydrogel lenses with oxygen transmissibility (Dk/t) of equal or more than 100 units and two non-silicone hydrogel lenses had oxygen transmissibility of 26 units or more, we were not expecting any lens-induced corneal swelling. According to Holden and Mertz[27] criteria, the critical Dk/t of a lens should be 24 units to be worn under open-eye conditions, avoiding corneal swelling. Also, the reduction in IOP was not correlated with the oxygen transmissibility.

From this study, it can be concluded that three months of soft CL wear reduces IOP and the reduction is associated with the lens materials and characteristics but not with the age and gender of the subjects. The reduction was positively correlated with corneal biomechanical properties and was higher during the first two months. Comfilcon A lenses reduced IOP more than the other lenses used. Eye care practitioners are recommended to consider the effect of CL wear before interpretation of IOP values measured in CL wearers. A long-term longitudinal study is suggested to confirm this finding.

References

1. Stapleton F, Keay L, Jalbert I, Cole N. The epidemiology of contact lens related infiltrates. *Optom. Vis. Sci.* 2007;84(4):257-72.

2. Ambroziak AM, Szaflik JP SJ. Therapeutic use of a silicone hydrogel contact lens in selected clinical cases. *Eye Contact Lens* 2004;30(1):63-7.

3. Zhang L, Chan O, Roy L, Barr JT. A meta-analysis of studies on cosmetically tinted soft contact lenses. *Clin. Ophthalmol.* 2013;7:2037-2042.

4. Garhwal R, Shady SF, Ellis EJ, Ellis JY, Leahy CD, McCarthy SP, Crawford KS GP. Sustained ocular delivery of ciprofloxacin using nanospheres and conventional contact lens materials. *Investig. Ophthalmlogy Vis. Sci.* 2012;53(3):1341-52.

5. Yao H, Shum AJ, Cowan M, Lähdesmäki I, Parviz BA. glucose level. *Biosens Bioelectron* 2011;26(7):3290-3296.

6. Hamilton KE, Pye DC, Hali A, Lin C, Kam P, Ngyuen T. The effect of contact lens induced corneal edema on Goldmann applanation tonometry measurements. *J. Glaucoma* 2007;16(1):153-8.

7. Steffen RB, Schnider CM. The impact of silicone hydrogel materials on overnight corneal swelling. *Eye Contact Lens* 2007;33(3):115-20.

8. Sweeney DF. Have silicone hydrogel lenses eliminated hypoxia? *Eye Contact Lens* 2013;39(1):53-60.

9. Ren DH, Petroll WM, Jester J V, Cavanagh HD. The effect of rigid gas permeable contact lens wear on proliferation of rabbit corneal and conjunctival epithelial cells. *CLAO J* 1999;25(3):136-41.

10. Pérez JG, Méijome JMG, Jalbert I, Sweeney DF, Erickson P. Corneal epithelial thinning profile induced by long-term wear of hydrogel lenses. *Cornea* 2003;22(4):304-7.

11. Harada Y, Hirose N, Kubota T, Tawara A. The influence of central corneal thickness and corneal curvature radius on the intraocular pressure as measured by different

tonometers: noncontact and goldmann applanation tonometers. *J. Glaucoma* 2008;17(8):619-25.

12. Saleh TA, Adams M, McDermott B, Claridge KG, Ewings P. Effects of central corneal thickness and corneal curvature on the intraocular pressure measurement by Goldmann applanation tonometer and ocular blood flow pneumatonometer. *Clin. Experiment. Ophthalmol.* 2006;34(6):516-20.

13. Cankaya AB, Beyazyildiz E, Ileri D OF. The effect of contact lens usage on corneal biomechanical parameters in myopic patients. *Cornea* 2012;31(7):764-9.

14. Ogbuehi KC, Osuagwu UL. Corneal biomechanical properties: Precision and influence on tonometry. *Contact Lens Anterior Eye* 2013.

15. Jorge J, González-Méijome JM, Queirós A, Fernandes P, Diaz-Rey JA. A comparison of the NCT Reichert R7 with Goldmann applanation tonometry and the Reichert ocular response analyzer. *Ophthalmic Physiol. Opt.* 2011;31(2):174-9.

16. Sudesh S, Moseley MJ, Thompson JR. Accuracy of Goldmann tonometry in clinical practice. *Acta Ophthalmol.* 1993;71(2):185-8.

17. Colás-Tomás T, Prieto-Del Cura M, Villafruela-Güemes I, Clariana-Martín A, Valdivia-Pérez A. [Comparison of dynamic contour tonometry, Goldmann and pneumotonometer in ocular hypertension patients and their relationship to pachymetry and ocular pulse amplitude]. *Arch. Soc. Esp. Oftalmol.* 2012;87(12):401-6.

18. Neuburger M, Maier P, Böhringer D, Reinhard T, Jordan J. The impact of corneal edema on intraocular pressure measurements using goldmann applanation tonometry, Tono-Pen XL, iCare, and ORA: an in vitro model. *J. Glaucoma* 2013;22(7):584-90.

19. Sapkota K, Franco S, Lira M. Intraocular pressure measurement with ocular response analyzer over soft contact lens. *Contact lens anterior eye* 2014;37(6):415-9.

20. Oliveira C, Franco S. Measuring Cornea. In: *Advance in Eye Research*.; 2012:115-36.

21. Fabian ID, Barequet IS, Skaat A, *et al*. Intraocular pressure measurements and biomechanical properties of the cornea in eyes after penetrating keratoplasty. *Am. J. Ophthalmol.* 2011;151(5):774-81.

22. Mahjoob M, Azimi A, Momeni-moghadam H, Mohamadian M, Yousofi R. The Effect of Various Contact Lenses on Intraocular Pressure Measurement by Goldman Tonometer. *Zahedan J Res Med Sci* 2014;16(6):33-35.

23. Oh JH, Yoo C, Kim YY, Kim HM SJ. The effect of contact lens-induced corneal edema on Goldmann applanation tonometry and dynamic contour tonometry. *Graefes Arch Clin Exp Ophthalmol* 2009;247(3):371-5.

24. Luce DA. Determining in vivo biomechanical properties of the cornea with an ocular response analyzer. *J. Cataract Refract. Surg.* 2005;31(1):156-62.

25. Touboul D, Roberts C, Kérautret J, *et al*. Correlations between corneal hysteresis, intraocular pressure, and corneal central pachymetry. *J. Cataract Refract. Surg.* 2008;34(4):616-22.

26. Bourne WM. The effect of long-term contact lens wear on the cells of the cornea. *CLAO J* 2001;27(4):225-30.

27. Holden BA, Mertz GW. Critical oxygen levels to avoid corneal edema for daily and extended wear contact lenses. *Invest. Ophthalmol. Vis. Sci.* 1984;25(10):1161-7.

28. Szczotka-Flynn L. Lens Distinctions. *Contact Lens Spectr.* 2007;22(June).

CHAPTER 7.

CONTRAST SENSITIVITY FUNCTION WITH SOFT CONTACT LENS WEAR

Highlights

- **Contrast sensitivity function (CSF) was measured before and after soft contact lens wear in neophyte subjects.**
- **CSF was found better with SCL than with spectacles.**
- **No change in CSF was obtained after three months of lens wear.**
- **No effect was seen with different lens materials and wearing modality.**

This chapter is based on the following original article: Sapkota K, Franco S, Lira M. Contrast sensitivity function with soft contact lens wear. Paper in the process of submission.

7.1 Introduction

Visual threshold to determine the smallest difference in contrast in between the visible and invisible is the threshold contrast, the reciprocal of which gives contrast sensitivity (CS).[1] Generally, sinusoidal gratings, which contain a gradual change from highest luminance to lowest luminance and vice-versa, are used to determine the visual contrast threshold. It is measured with different spatial frequencies and with different contrasts, so it measures the real-world visual function, unlike the visual acuity measurement. Visual acuity only indicates the visual performance on high contrast, generally it is measured in 100% contrast target. It can not be a complete tool to represent the visual performance because contrast greatly varies in the real world visual requirements and wide range of targets have about 1% of contrast. In some conditions of eyes, contrast sensitivity may be reduced significantly even visual acuity is normal.[1]

Many researchers included CS test to determine the visual performance with different types of contact lenses (CL). Some have evaluated contrast sensitivity as the function of different spatial frequencies, that is contrast sensitivity function (CSF),[2-4] while others have measured visual acuity with high and low contrast visual acuity charts.[5,6] However, low contrast visual charts reflect only the slope of higher spatial frequency level.[7] Some of the previous studies found no difference in CS with CL and without correction,[6] but a couple of studies showed that CS decreases with CL.[2,4,8] Porish showed that CS can be improved in athletes with sport-tinted CL.[9] The author found enhancement of CS with sport-tinted CL but no change in CS with clear CL. However, although it was statistically significant, the improvement was too small to affect the performance of the athletes.

As far as the authors know, none of the previous studies have determined the effect of contemporary CL wear on CSF. The aim of this study was to investigate the effect of three months of soft CL wear in CSF. CSF with CL during baseline was compared with spectacles; and CSF with CL after three months of lens wear. The effect of lens materials, wearing modality, age and gender on CS was also evaluated.

7.2 Methods

Ninety-four eyes of 45 healthy subjects (18 years or older) without ocular pathology were included in this longitudinal clinical trial conducted in University of Minho, Portugal. After detailed information provided to the subjects, they signed a consent form. This study followed the tenets of Declaration of Helsinki and ethical approval was provided by the Ethical committee of the School of Sciences. All the subjects had myopic refractive error with best corrected visual acuity equal or better than 6/6 in each eye. Subjects with astigmatism more than 0.75D and past history of CL wear were excluded. The sample size was computed on the basis of preliminary data to warrant 80% statistical power with 0.05 significant level to detect the difference in LogCS by 0.15 between two conditions: with spectacles and CL at baseline; and with CL at baseline and after three months of wear.

Preliminary ophthalmological examination was done to select the candidates from all the people willing to participate in the study. Visual acuity was measured in each subject without any visual correction and with spectacles. CSF was measured with CSV-1000 (VectorVision, Greenvile, OH).[3,10,11] It is one of the commercially available CS measurement tools with good reliability which applied sinusoidal gratings.[12] It applies objective method by forced-choice detection[1] and measures the CS in four levels of spatial frequencies: 3, 6, 12 and 18 cycles per degree (cpd). Each set of spatial frequency contains 17 patches with diameter 3.8 cm or 0.87 degree. The first patch with highest contrast vertical sinusoidal gratings is for explanation while other 8 pairs of patches, numbered 1 to 8, are for testing. Each pair contains one with vertical sinusoidal gratings and the next plane and these are randomly located either upside or downside of the column. Contrast of the 1st patch to 3rd decreases in 0.17 log unit steps while from 3rd to 8th decrease in 0.15 log unit steps. The chart is retro-illuminated with 85 cd/m^2 fluorescent light which can be controlled by a remote. Subject was seated two and a half meters away from the chart. CSF was measured in each eye separately with spectacles. CSF of the last correct response was recorded. Because sensitivity is the inverse of contrast values, log unit was used so that higher log values indicates better sensitivity.[13] Each sinusoidal grating patch contains a bar with the brightest luminance and darkest luminance, the thickness of the bar depends upon the spatial frequency of the gratings, e.g. for 3 cpd it is wider in comparison to that of 18 cpd. Thus,

$$Contrast\ threshold = \frac{Luminance\ Maximum - Luminance\ Minimum}{uminance\ Maximum - Luminance\ Minimum} \quad (7.1)$$

Table 7-1. Characteristics of the lenses used in the study

	Lotrafilcon B	**Nelfilcon A**	**Comfilcon A**	**Stenofilcon A**	**Balafilcon A**	**Nesofilcon A**
Company	Alcon	Alcon	Cooper Vision	Cooper Vision	Bausch & Lomb	Bausch & Lomb
Brand name	AirOptix® Aqua™	Dailies® AquaComfort Plus	Biofinity™	MyDay™	Purevision2™	Biotrue™ ONEday
Water content (%)	34	69	48	54	36	78
Thickness (mm)	0.08	0.10	0.08	0.08	0.07	0.1
Base curve/ diameter (mm)	8.6/14.2	8.7/14	8.7/14.5	8.4/14.2	8.6/14	8.6/14.2
Oxygen Permeability (barrer)	110	26	128	80	91	42
Modulus (MPa)	1.2	0.89	0.75	0.4	1.1-1.25	0.49
Transmisibility (barrer/cm)	137.5	26	160	100	130	42

Tear function test, keratometry, biomicroscopic examination and pre-contact lens evaluation were done in each subject. Subjects were fitted with CL in such a way that one eye was fitted with a daily disposable lens (Nelfilcon A or Stenofilcon A or Nesofilcon A) and the other was fitted with a monthly disposable lens (Lotrafilcon B or Comfilcon A or Balafilcon A) in a contralateral manner. Details about CL are presented in table 7.1. CSF was again

measured with CL within 30 minutes of lens wear. Subjects were again evaluated including the CSF assessment, after three months of CL wear.

Data were analyzed with Statistical Software for Social Sciences (SPSS 22, IBM Corp., Armonk, NY). Descriptive variables were expressed in mean ± standard deviation (SD). Kolmogorov-Smirnov test was done to analyze the distribution of the variables; parametric tests were used to test normally distributed variables and non-parametric tests were applied to others. CSF expressed in Log values in different spatial frequencies with spectacles and with CL were compared with Wilcoxon signed rank test. The correlation of changes in contrast sensitivity with age was determined by Spearman's rho test. Association of changes in CSF with gender and type of lenses was assessed by Mann-Whitney test. P values less than 0.05 were considered as statistically significant.

7.3 Results

Ninety-four eyes of 47 subjects with mean age 24.3± 4.1 years were included in this study. Among them, 66.0% (31) were female. Lotrafilcon B was worn in 16 eyes, Nelfilcon A on 16 eyes, Comfilcon A on 15 eyes, Stenofilcon A on 15 eyes, Balafilcon A on 16 eyes and Nesofilcon A on 16 eyes.

Values of CSF before starting to wear CL (with spectacle correction), with CL during the first day of lens wear and with CL after three months of lens wear in different spatial frequencies are shown in table 7.2.

Table 7-2. Contrast sensitivity function (LogCS) with spectacles and with contact lenses

Spatial frequency (cpd)	Baseline with spectacles	Baseline with CL	p*	Final with CL	p**
3	1.58±0.20	1.66±0.16	0.001	1.67±0.14	0.502
6	1.76±0.23	1.86±0.20	0.000	1.88±0.19	0.316
12	1.44±0.26	1.53±0.24	0.004	1.52±0.25	0.875
18	1.03±0.28	1.08±0.23	0.114	1.09±0.26	0.687

*comparison between CL and spectacles, ** comparison between baseline with CL and final with CL [CL: contact lenses; cpd: cycles per degree].

CS with CL was significantly higher than that of with spectacles wear during baseline evaluation in all the spatial frequencies ($p < 0.05$) except spatial frequency of 18 cpd, in which CS was similar with spectacles and CL ($p = 0.114$). In average, CSF with CL was 0.07 higher than the CS values with spectacles. But there was not a significant change in CSF with CL during baseline examination in comparison to the values after three months in each frequency level ($p > 0.05$). As shown in figure 7.1, CS was higher with a medium level of spatial frequency in comparison to lower or higher spatial frequencies.

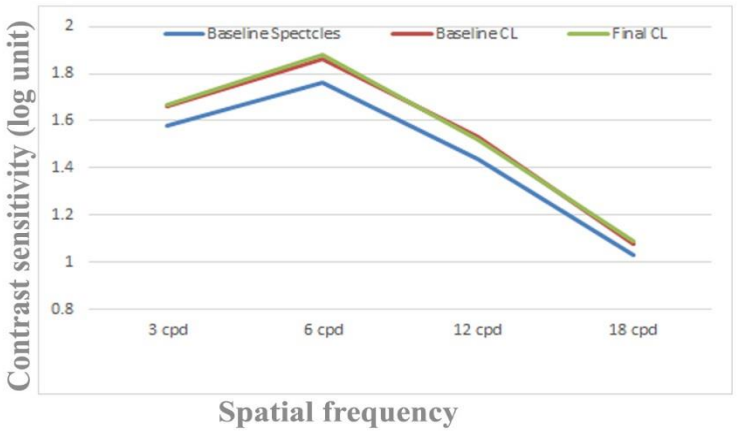


Figure 7.1. Graph showing CSF with different spatial frequencies.

On table 7.3 it is presenting the changes in CS function for the different materials of CL.

Changes in CSF were not correlated with age and gender of the subjects ($p > 0.05$). Figures 7.2 (a, b) show CSF of different lens materials with CL during baseline and on the final follow-up visit. Here, different materials behaved differently; however, the difference was not statistically significant ($p > 0.05$). Change in CSF was not correlated with the refractive power of the lenses ($p > 0.05$).

Table 7-3. Change in contrast sensitivity function for different spatial frequencies with different lens materials

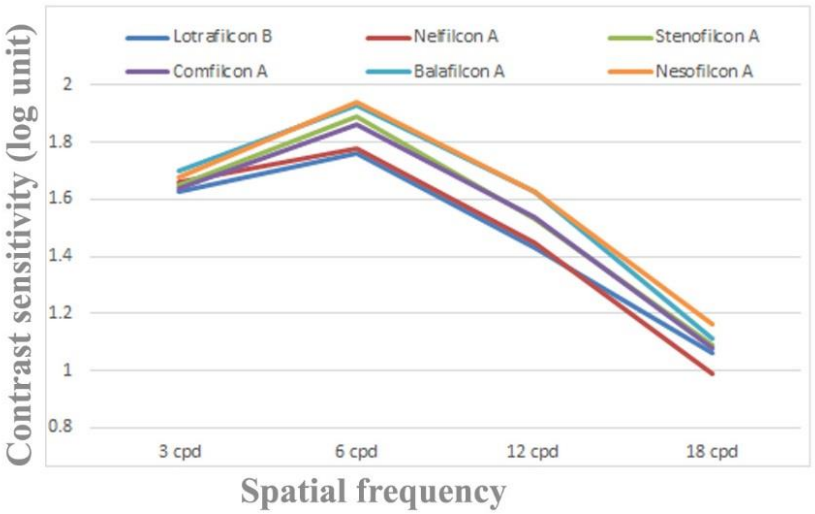
	Frequency			
	3 cpd	**6 cpd**	**12 cpd**	**18 cpd**
Lotrafilcon B	0.04±0.17 [0.464]	0.08±0.22 [0.217]	0.07±0.40 [0.608]	-0.02±0.31 [0.805]
Comfilcon A	0.04±0.09 [0.052]	-0.01±0.18 [0.320]	0.05±0.26 [0.506]	0.003±0.24 [0.758]
Balafilcon A	-0.01±0.15 [0.464]	0.001±0.17 [0.916]	-0.13±0.27 [0.053]	0.02±0.27 [0.959]
Nelfilcon A	-0.02±0.19 [0.465]	0.05±0.22 [0.422]	0.03±0.33 [0.527]	0.07±0.29 [0.318]
Stenofilcon A	0.02±0.10 [0.330]	0.03±0.17 [0.354]	0.01±0.13 [0.892]	-0.04±0.18 [0.473]
Nesofilcon A	0.03±0.18 [0.549]	-0.01±0.12 [0.863]	-0.07±0.33 [0.717]	0.04±0.28 [0.474]

Mean values ± standard deviation are followed by p values in bracket.
cpd: cycles per degree

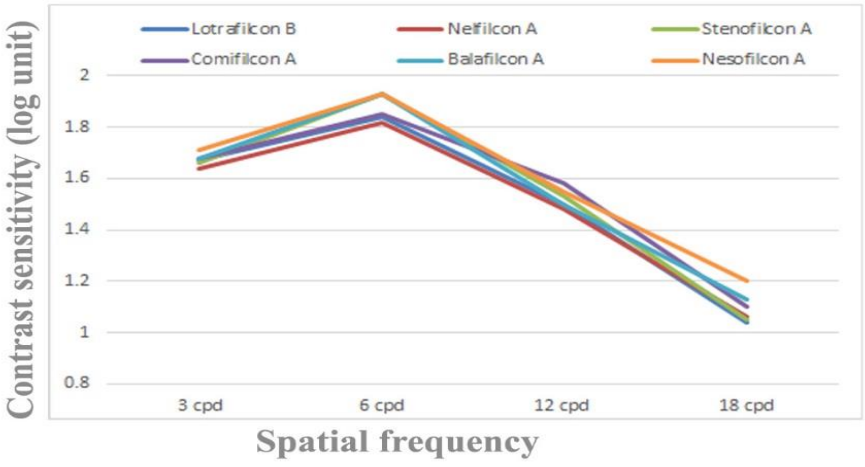
7.4 Discussion

Traditionally, visual performance with soft CL is most frequently measured with high-contrast visual acuity measured with the Snellen acuity charts. This study measured the effect of soft CL wear in CSF comparing the scores obtained with spectacles and after three months of lens wear. The effect of lens materials, wearing modality, age and gender were also evaluated. It was found significantly higher CSF with CL in comparison to the CSF with spectacles on the first examination day in all the spatial frequencies except one spatial frequency of 18 cpd. The lower the spatial frequency, the higher was the improvement. Similar to our findings, Dalcoll *et al*. found an increase in CSF with soft CL.[14] CSF is the product of both neural and optical factors.[1] Neurally, different channels are selective to different spatial frequencies. Aberration, glare affect the quality of an image. So the improvement in CSF with CL in comparison to spectacles may be due to the smaller asymmetric aberrations with soft

CL.[15] Charman[16] reported that incorrect refractive error correction reduces the CSF in high spatial frequency. So, in the current study, no difference in CSF with spectacles and CL in high spatial frequency may be due to uncorrected astigmatism with spherical CL. In the current study, the difference in CSF was less than 0.14 Log units in all the spatial frequencies, so this difference may not be clinically significant.[17]



a



b

Figure 7.2. Contrast sensitivity function with different spatial frequencies a. baseline with contact lenses; b. final with contact lenses.

Contrary to the current study, Barth *et al*. did not find any difference in CSF with spectacles and with soft CL (Acuvue 2, Biomedics 55 and Focus 1-2 week).[18] Wachler *et al*.[19] did not find any significant difference in the CSF with CL and spectacles with Acuvue and Biomedics, except Cibasoft material, with which CSF was less in comparison to spectacles in 12 cpd spatial frequency. However, they had a small number of subjects (20 eyes of 10 subjects) and different lens material was used. Belda-salmeron did not find changes in CSF by soft lens wear in comparison to the CSF without correction.[6] Gray suggested that changes in corneal physiology due to CL wear may reduce the CSF.[20,21] However, in the current study CSF was measured with CL within half an hour of lens wear. So in this short period, there might not be any change in corneal physiology. Carracedo *et al*[22] found an increase in CS with filter CL in retinitis pigmentosa patients. CS was better with CL than with spectacles.

Another important finding of the current study was that three months of soft CL wear did not change the CSF. This highlights the fact that contemporary soft CL wear does not damage corneal physiology so that no effect was observed. Soni *et al.* suggested that CL with higher thickness and low oxygen transmissibility may reduce CS by inducing corneal edema.[7] But in the current study, CL were worn for daily wear modality and the central thickness of the lenses was not more than 0.10mm. The oxygen transmissibility of CL was enough to avoid the corneal edema on open eye condition.[23] Supporting our findings, Grey found no change in CSF by six months of soft CL wear.[21] In his six-sample size study, he concluded that CSF decreases by soft CL wear initially, but it rapidly recovers to a normal state within few hours. In another study, he found a reduction in CSF with soft CL after few hours of wear in comparison to the initial CSF without any correction.[20] Belda-Salmeron *et al*. did not find any change in CSF just after the lens wear and up to 12 hours of lens wear.[6] However, their findings may not be comparable with the current study because they used different types of lenses and applied different time frames.

As shown in figure 7.2, CSF varied with lens materials. There was a smaller difference in CSF on lower spatial frequency in comparison to the CSF on higher spatial frequencies and this variation was highly perceivable after three months of CL wear. However, this variation was not statistically significant. Contrary to the present findings, Belda-Salmeron found that CSF depends upon the lens materials as well as lens design and manufacturing methods.[6] They

reported a severe reduction in CSF with thicker lenses than thinner lenses and found better CSF with lathe-cut lenses than with spectacles or cast-molded lenses. Gupta *et al*. compared the CSF with two types of CLs, Purevision multifocal and Purevision single vision lenses, and found no difference in CSF.[24] Similarly, Fernandes *et al*. did not find any significant difference in CSF with multifocal or monovision CL in presbyopic subjects.[5]

This study applied CSV-1000 for the contrast sensitivity measurement. Some of the studies found lower reliability of CSV-1000 in CS measurement.[25] However, the same test was used during all the visits in this longitudinal study and the same examiner measured all the data. It was reported that reliability increases if performed by the same examiner.[25]

Thus, from this study it can be concluded that CSF improves with soft CL in comparison to the CSF with spectacle. Wearing CL neither reduces nor increases the CSF. CSF with CL was independent of the lens materials, designs of the lenses and the age and gender of the subjects. Studies using toric and multifocal lenses with varied thickness would be more interesting to know the effect of CL wear on CSF in future.

References:

1. Pelli DG, Bex P. Measuring contrast sensitivity. *Vision Res.* 2013;90:10-14.

2. Briggs ST. Contrast sensitivity assessment of soft contact lens wearers. *Int. Contact Lens Clin.* 1998;25(98):99-102.

3. Dorn L. Contrast Sensitivity in Strabologic Functional Test. *Acta Clin Croat* 2008;47(suppl 1):15-20.

4. De Fez MD, Luque MJ, Viqueira V. Enhancement of contrast sensitivity and losses of chromatic discrimination with tinted lenses. *Optom. Vis. Sci.* 2002;79(9):590-597.

5. Fernandes PRB, Neves HIF, Lopes-Ferreira DP, Jorge JMM, González-Meijome JM. Adaptation to multifocal and monovision contact lens correction. *Optom. Vis. Sci.* 2013;90(3):228-235.

6. Belda-Salmerón L, Ferrer-Blasco T, Albarrán-Diego C, Madrid-Costa D, Montés-Micó R. Diurnal variations in visual performance for disposable contact lenses. *Optom. Vis. Sci.* 2013;90(7):682-90.

7. Soni PS, Patel R, Carlson RS. Is binocular contrast sensitivity at distance compromised with multifocal soft contact lenses used to correct presbyopia? *Optom. Vis. Sci.* 2003;80(7):505-514.

8. Ortiz C, Jimenez R. Optical Quality and Vision with Iris-Coloring Soft Contact Lenses. *Optom. Vis. Sci.* 2014;91(5):564-569.

9. Porisch E. Football players' contrast sensitivity comparison when wearing amber sport-tinted or clear contact lenses. *Optometry* 2007;78:232-235.

10. Sia DIT, Martin S, Wittert G, Casson RJ. Age-related change in contrast sensitivity among Australian male adults: Florey Adult Male Ageing Study. *Acta Ophthalmol.* 2013;91:312-317.

11. Franco S, Silva AC, Carvalho AS, Macedo AS, Lira M. Comparison of the VCTS-6500 and the CSV-1000 tests for visual contrast sensitivity testing. *Neurotoxicology* 2010;31(6):758-761.

12. Pomerance GN, Evans DW. Test-retest reliability of the CSV-1000 contrast test and its relationship to glaucoma therapy. *Investig. Ophthalmol. Vis. Sci.* 1994;35(9):3357-3361.

13. Campbell FW, Robson JG. Application of Fourier analysis to the visibility of gratings. *J. Physiol.* 1968;197:551-566.

14. Dalcoll MW, Alves MR, Barreto J, Yamane IDS, Bechara S, Mukai A. Evaluation of optical performance of soft contact lenses in myopic correction. *Arq. Bras. Oftalmol.* 2008;71(6 suppl):37-41.

15. Hong X, Himebaugh N, Thibos LN. On-Eye Evaluation of Optical Performance of. *Optom. Vis. Sci.* 2001;78(12):872-880.

16. Charman WN. Effect of refractive error in visual tests with sinusoidal gratings. *Br. J. Physiol. Opt.* 1979;33(2):10-20.

17. Arditi A, Cagenello R. On the statistical reliability of letter-chart visual acuity measurements. *Investig. Ophthalmol. Vis. Sci.* 1993;34(1):120-129.

18. Barth B, Alves MR, Kara-José N. Visual performance in myopic correction with spectacles and soft contact lenses. *Arq. Bras. Oftalmol.* 2008;71(1):90-96.

19. Wachler BS, Phillips CL, Schanzlin DJ, Krueger RR. Comparison of contrast sensitivity in different soft contact lenses and spectacles. *Clao J* 1999;25:48-51.

20. Grey CP. Changes in contrast sensitivity during the first hour of soft lens wear. *Am. J. Optom. Physiol. Opt.* 1986;63:702-707.

21. Grey CP. Changes in contrast sensitivity during the first six months of soft lens wear. *Am. J. Optom. Physiol. Opt.* 1987;64:768-774.

22. Carracedo G, Carballo J, Loma E, Felipe G, Cacho I. Contrast sensitivity evaluation with filter contact lenses in patients with retinitis pigmentosa: A pilot study. *J. Optom.* 2011;4(4):134-139.

23. Holden B, Mertz GW. Critical oxygen levels to avoid corneal edema for daily and extended wear contact lenses. *Invest. Ophthalmol. Vis. Sci.* 1984;25(10):1161-7.

24. Gupta N, Naroo S a, Wolffsohn JS. Visual comparison of multifocal contact lens to monovision. *Optom. Vis. Sci.* 2009;86(2):E98-E105.

25. Kelly S., Pang Y, Klemencic S. Reliability of the CSV-1000 in Adults and Children. *Optom. Vis. Sci.* 2012;89(8):1172-1181.

CHAPTER 8.

CONCLUSION, LIMITATION AND FUTURE WORK

8.1 Conclusions

The work presented in this thesis obtained several important information regarding the effect of SCL wear on the ocular surface. With the experiments explained in this thesis, it was also acquired information about the effect of different lens materials and wearing modality on the changes occurred by lens wear which can help to better predict the response of each type of lens. The main conclusions developed from this thesis are summarized as follow:

- SCL wear affects conjunctival cytology. It was found a significant reduction in goblet cell density (GCD) after three months of lens wear. So, since mucin is secreted by goblet cells and has an important role in tear function, the etiology of the dry eye symptom present in SCL wearers may be the loss of GCD. Although there is a high variation in GCD and this fact does not allow to estimate the exact degree of GCD reduction, since this study was conducted in neophyte CL wearers, the reduction observed might be sole effect of CL wear. CL wear can induce physical, as well as mechanical irritation on the ocular surface since continuous movement of the lens due to blink, may cause friction on the conjunctival surface affecting goblet cells. This effect of lens wear on goblet cells was associated with lens materials but not with lens care system. Hydrogel contact lenses (HCL) induced more effect than SiHy lenses and this may be due to the fact that HCL produces more irritation or HCL wear does not respect the ocular physiology because of their lower oxygen transmissibility. The effect was maximum with Nelfilcon A lens wear while Comfilcon A did not affect GCD. In our experiment, epithelial cell metaplasia was increased by at least one grade in the majority of the eyes, still it was not statistically significant. This experiment highlights that goblet cells are more affected by lens wear than epithelial cell morphology. Interestingly, there was not a significant difference in changes in conjunctival cytology between daily disposable and monthly disposable lens wear.
- SCL induced significant changes on ocular surface physiology as conjunctival limbal and bulbar redness, conjunctival and corneal staining were increased with lens wear. Conjunctival redness, specifically limbal redness is an indicator of hypoxia with CL wear and the changes observed even with hyper permeable SiHy lenses were surprising.

Since oxygen transmissibility (Dk/t) on the thicker peripheral part of the myopic lenses is very different when compared with the central Dk/t, it is probably insufficient to avoid hypoxic effect in the peripheral cornea or limbus. Conjunctival redness was associated with the power of the lenses with higher power myopic lenses inducing higher effects. Conjunctival redness was higher in the temporal and nasal part which may be due to the higher tear evaporation on these exposed parts. The important finding of this experiment was that the changes on ocular surface physiology were higher during the early period of lens wear and minimum on latter period. Bulbar redness and corneal staining were also associated with lens materials. Again, no significant difference was observed in changes in ocular surface physiology with wearing modality of the lenses.

- SCL wear affects some of the corneal biomechanical properties. In the present study, corneal resistance factor (CRF) was reduced with three months of SCL wear. Although statistically non-significant, we found flattening of the anterior corneal curvature. So, reduction in CRF may be due to changes in corneal curvature. Reduction in CRF was associated with lens materials. It was maximum with Comfilcon A wear and negligible with Balafilcon A, Nesofilcon A and Lotrafilcon B lens wear. Even SiHy lenses behaved differently which may be due to their different properties. During the same period of lens wear, corneal hysteresis (CH) remained same.

- Our experiment shows that measurement of intraocular pressure (IOP) over SCL with Ocular Response Analyzer (ORA) underestimates the IOP. This underestimation may be due to the reduction in the time needed to achieve the maximal light detection in non-contact tonometry when the front surface is flattened by an air puff. The majority of the eyes had the mean underestimation less than 3 mmHg which is considered as the acceptable values, but to get the accurate values CL should be removed. If the measurement with ORA has to be done over CL, Goldmann-correlated intraocular pressure should be used which is less affected in comparison to corneal-compensated intraocular pressure.

- After three months of SCL wear in a daily wear basis, IOP was reduced. Reduction in IOP was higher with cornea having higher CRF which can indicate that IOP reduction

is lower in a fragile cornea. The cause of the reduction in IOP may be multifactorial and in the present study it was associated with the lens material.

- Our research found that contrast sensitivity function (CSF) with CL is higher than with spectacles for all the lower spatial frequencies. Since CSF is the product of both neural and optical factors, CL wear may reduce asymmetric aberrations and improve CSF. However, the difference in CSF obtained with spectacles and CL was less than 0.14 Log unit which is considered as clinically non-significant. There was no statistically significant difference in CSF for the different lens materials or different wearing modality. Three months of SCL wear did not change CSF.

8.2 Limitations

It was tried to make this thesis out of error and limitations. However, due to some uncontrolled conditions, there are some limitations/delimitations in the experiments. Next it is summarized important limitations of the studies which could affect the results.

1. The first experiment was conducted on the conjunctival cytology. It was performed the impression on the superior bulbar conjunctiva and the findings may not represent the GCD and epithelial cell morphology of the whole conjunctiva.
2. Changes on ocular surface physiology were graded by a subjective method which may cause some human error or bias in comparison to the objective grading.
3. It was investigated the effect of SCL wear on corneal biomechanical properties and was found a reduction in CRF but no change in CH. Indirectly, our experiments showed that there was not corneal swelling or change in corneal thickness and it could be more interesting if it was measured corneal thickness and correlate it with corneal biomechanical properties. The same situation applies with the results obtained in IOP.
4. Out of the seven studies presented in this thesis, five were longitudinal studies. The variables were compared between the final and the baseline visit. So, the baseline data can be assumed as the control. However, if it was included a different group of subjects as a control group, the results would be less bias but other factors would be missed.
5. This thesis presented trials conducted in a contralateral design, with different types of lenses worn in right and left eyes. It was assumed that all subjects wore lenses in the

proper way. So the incorrect use of the lenses could cause error on the results. However, to ascertain the proper use of lenses, a paper was provided where they should note which lens was worn on right eye and which one on left eye.

6. Due to the nature of the study—a Ph.D. thesis-- all the data were collected by a single investigator and this might induce some bias in such unmasked study.

8.3 Future works

From the discussion and the conclusions of this thesis, some new questions have been emerged that should be addressed in the future using the knowledge acquired from the series of experiments presented in this thesis.

1- It was performed a cytological examination on the superior bulbar conjunctiva and significant changes were observed. GCD reduced and epithelial cell metaplasia grading increased. From these findings, new questions have been raised. What will be the effect of long-term SCL wear? Does the changes in conjunctival cytology continue or come to a normal state after some period? What is the effect on other parts of the conjunctiva? So, future cytological study with long-term CL wear is suggested.

2- Evaluation of conjunctival and corneal physiology with an objective method may give less bias results. Moreover, use of the greatest score of the different parts of the cornea or conjunctiva rather than the average score of grading may give more clinically important results. Such studies are suggested for the future.

3 - CRF was found to be reduced by three months of SCL wear. More studies with long-term CL wear are necessary to determine whether such changes are temporary or permanent.

4 -Three months of SCL wear reduced IOP. The reduction in IOP was higher initially and less in later period. Assessing whether this condition is reversible or not should be analysed and it is an important question raised from this study. To address this question, a long-term study is necessary.

APPENDIX I

ABSTRACTS PRESENTED IN CONFERENCES

Intraocular pressure measurement with Ocular Response Analyzer over soft contact lens. [Congresso Internacional de Optometria e Ciências da Visão (CIOCV) 2013]

Objectives: Measurement of intraocular pressure is useful in patients having risk factors of glaucoma. We aim to compare intraocular pressure measured with Ocular Response Analyzer (ORA) with and without contact lenses (CL) on the eye.

Methods: Goldmann-correlated intraocular pressure (IOPg) and corneal-compensated intraocular pressure (IOPcc) were measured in 56 eyes of 28 subjects without any ocular pathology using ORA. One eye was fitted with Narafilcon A (1-Day Acuvue True Eye, Johnson & Johnson) and the other eye with Nelfilcon A (Daily AquaComfort Plus, Ciba Vision) randomly and IOPg and IOPcc were again measured over CL. The variation in the intraocular pressure with and without CL was determined.

Results: Out of 28 subjects, 53.6% (15) were female. The mean age of the subjects was 29.39±9.84 years with range 20 to 51 years. Both the IOPg and IOPcc when measured with CL were found statistically significantly lower than without CL ($p < 0.05$). In subjects wearing Narafilcon A lens, IOPg and IOPcc were found 0.88 ±2.04 mmHg and 1.55±2.16 mmHg lower than without CL, respectively. Similarly, with Nelfilcon A lens, IOPg and IOPcc were found to be 1.03±1.93 mmHg and 1.62±3.12 mmHg lower, respectively. The difference was higher with Nelfilcon A lens, however, it was statistically not significant. The variation in the IOPg and IOPcc with and without lenses was not associated with age, gender or race ($p > 0.05$).

Conclusion: ORA showed statistically significant lower intraocular pressure when subjects are wearing Narafilcon A and Nelfilcon A soft CL in comparison to pressure without lenses. However, these differences can be considered clinically insignificant and when IOP is necessary to measure in CL wearers, it might be measured with CL on eye.

Key words: Ocular response analyzer, corneal compensated intraocular pressure, soft contact lens.

Use of ORA to assess mechanical properties of soft contact lenses: a pilot study [CIOCV 2013]

Introduction: Ocular biomechanical properties have applications in a variety of important areas including refractive surgery, corneal disease and glaucoma and can be measured with Ocular Response Analyzer (ORA). With this study, we aim to investigate if the same instrument can be used to give information about contact lenses mechanical properties.

Methods: It was a prospective, cross-sectional study. Twenty-eight subjects with normal eyes were recruited. Corneal hysteresis (CH) and corneal resistance factor (CRF) were measured using ORA. Each subject was fitted with a silicone hydrogel lens (1-Day Acuvue True Eye, Johnson & Johnson) in one eye and a hydrogel lens (Dailies AquaComfort Plus, Ciba Vision) in fellow eye in a random manner and CH and CRF were re-measured with contact lens on. The variations of these properties with and without contact lens were analyzed.

Results: The mean age of the subjects was 29.39±9.84 years with range 20-51 years. CH was found to be higher with the contact lens in comparison to without contact lens, but it was statistically significant only with the silicone hydrogel lens ($p = 0.019$). CH was 0.73±1.55mmHg and 0.68±1.98 mmHg higher with silicone and nonsilicone hydrogel lenses, respectively. CRF was found to be 0.36±1.57 mmHg and 0.27±1.60 mmHg higher with silicone and nonsilicone lens respectively, but none of them were statistically significant ($p > 0.05$). The variation in the ocular biomechanical properties was not associated with age, gender and race ($p> 0.05$).

Conclusion: This study shows good consistency when CH and CRF are measured with ORA over hydrogel contact lens. The same was not observed for silicone-hydrogel lens probably due to its lower water content and higher modulus. This can be understood as an indirect measure of mechanical properties of contact lenses.

Key words: Corneal hysteresis, corneal resistance factor, soft contact lens.

Changes in conjunctival redness and corneal staining by soft contact lens wear [American Academy 2014, Denver]

Purpose: To determine the changes in conjunctival bulbar/limbal redness and corneal staining by soft contact lens wear.

Methods: A longitudinal clinical trial was conducted in normal myopic subjects who had never worn contact lens before. Subjects were fitted with a monthly or daily lens, Lotrafilcon A or Nelfilcon A or Comfilcon A or Stenofilcon A on contra-lateral eyes randomly. Conjunctival bulbar/limbal redness and corneal staining were evaluated before and after three months of wearing contact lenses.

Results: Forty-two eyes of 21 subjects were included in this study with mean age 23.5 ± 3.5 years. After three months of soft contact lens wear, bulbar redness ($p < 0.001$), limbal redness ($p < 0.001$) and corneal staining ($p < 0.001$) increased.

For both the type of lenses, bulbar redness increased during the first month ($p < 0.001$) and second month ($p = 0.001$) but then remained same ($p = 0.615$). Similarly, limbal redness increased during first month ($p = 0.001$) and second month ($p = 0.001$) but remained same during third month ($p = 0.078$). However, corneal staining was found increased during first two months ($p < 0.001$) and also during the third month ($p = 0.012$).

Grossly, there was not significant difference in conjunctival redness and corneal staining on different lens materials ($p > 0.05$), however, Comfilcon A lens produced significantly higher bulbar as well as limbal redness in comparison to other materials ($p<0.05$). Bulbar as well as limbal redness was found higher in nasal and temporal side and lower in superior and inferior side. Highest corneal staining was found on inferior area.

Conclusion: Soft contact lenses increase conjunctival redness and corneal staining. The changes are higher during early months and smaller on latter period.

Key words: Bulbar redness, corneal staining, soft contact lens

Three month soft contact lens wear changes conjunctival cytology [American Academy 2014, Denver]

Purpose: Contact lens related dry eye, which is a major problem in modern contact lens practice, might be correlated with alterations in ocular surface cytology. Thus the aim of this study was to investigate the changes in conjunctival cytology and to determine the changes in goblet cell density after soft contact lens wear for a period of three months.

Methods: Seventeen normal subjects who had never worn contact lenses before were fitted with a monthly or daily lens, Lotrafilcon A or Nelfilcon A or Comfilcon A or Stenofilcon A in contra-lateral eyes, randomly. Conjunctival impression cytology was performed before contact lens wear and after three months. Nitrocellulose filter paper was impressed on superior bulbar conjunctiva, fixed with ethanol and stained with periodic acid Schiff and Haemotoxyline/Eosin. Epithelial cell morphology, as well as goblet cell density, was analyzed on light microscope.

Results: Thirty-four eyes of 17 subjects were included in this study. Two-thirds of the subjects were females and mean age of the subjects was 23.1±3.1 years. There was no significant change in epithelial cell morphology induced by contact lens wear ($p = 0.059$). However, goblet cell density decreased significantly after three months of lens wear ($p< 0.001$). The reduction on the goblet cell density was observed with all the different contact lenses tested. Reduction in goblet cell density was not related with age, gender and lens materials ($p > 0.05$).

Conclusion: Soft contact lens wear did not alter epithelial cell morphology; however, the goblet cell density reduced significantly regardless the contact lens used. In conclusion we can say that reduction in goblet cells may contribute to contact lens related dry eyes.

Key words: Conjunctival impression cytology, goblet cell density, soft contact lens

Relationship of goblet cell density with tear function tests and ocular surface physiology [CIOCV 2014]

Objectives: Goblet cells are the main source of tear mucin which has an important role both for the optical properties of cornea as well as ocular comfort. The objective of this study was to determine the relationship of goblet cell density (GCD) with tear function tests and ocular surface physiology.

Methods: This was a cross-sectional clinical trial conducted in 70 eyes of 35 normal subjects with mean age of 24±3.7 years. Tear film assessment, and conjunctiva and cornea examination was done in each subject. Conjunctival impression cytology was done by applying Nitrocellulose Millipore MF^{TM}-Membrane filter over the superior bulbar conjunctiva. The filter paper was then fixed with 96% ethanol and stained with Periodic Acid Schiff. Goblet cell density was determined with the light microscope. Relation between GCD and Schirmer score, non-invasive tear break-up time (NIBUT), tear break-up time (TBUT), bulbar redness, limbal redness and corneal staining were determined.

Results: GCD was found higher in eyes with higher Schirmer score and NIBUT but there was not any significant relationship between GCD with Schirmer score ($p = 0.686$) and NIBUT ($p = 0.099$). However, there was a significant relationship of GCD with TBUT ($p = 0.042$), bulbar redness ($p = 0.003$), limbal redness ($p = 0.001$) and corneal staining ($p < 0.001$). No difference in GCD was found between women and men ($p = 0.564$).

Conclusion: GCD does not have any relationship with the aqueous portion of the tear. However, it is positively correlated with TBUT. GCD also correlated with limbal as well bulbar redness and corneal staining.

Key words: Goblet cell density, Schirmer score, tear break time, bulbar redness, corneal staining.

Effect of soft contact lenses wear on intraocular pressure [CIOCV 2014]

Objectives: Contact lenses are one of the main options of refractive error correction. The aim of this study was to determine the effect of soft contact lenses wear on intraocular pressure in normal eyes.

Methods: This was a longitudinal interventional clinical trial conducted in University of Minho, Portugal. Intraocular pressure was measured by Ocular Response Analyser in 24 normal eyes of 12 subjects who had never worn contact lenses before. Subjects were fitted with daily disposable Nelfilcon A or monthly disposable Lotrafilcon B contact lens in contra-lateral manner. Measurements of intraocular pressure were repeated every month during three months of contact lenses wear. Changes in Goldmann-correlated intraocular pressure (IOPg) were analyzed. Effect of corneal biomechanical properties [corneal resistance factor (CRF) and corneal hysteresis (CH)] on the changes of IOPg was also studied.

Results: IOPg reduced significantly after three months of contact lenses wear ($p = 0.01$). During the first month of wear, IOPg reduced significantly ($p = 0.029$) and continued to decrease in the second month also but the changes were not significant ($p = 0.68$). However, after that, it started to increase but with no significance (0.126). The changes in IOPg was not correlated with the lens materials ($p = 0.876$). CRF was found positively correlated with the changes in IOPg ($p = 0.013$) but CH was not ($p = 0.248$).

Conclusion: Soft contact lens reduces the IOPg during three months of lens wear in neophyte contact lens wearers. Study with larger sample size and longer duration is necessary to confirm this finding.

Key words: Goldmann-correlated intraocular pressure, corneal resistance factor, soft contact lens, corneal hysteresis

Effect of soft contact lens wear on corneal biomechanical properties and corneal topography [American Academy 2015, New Orleans]

Purpose: The aim of this study was to investigate the effect of soft CL wear on corneal hysteresis (CH) and corneal resistance factor (CRF). Effect of corneal topographic indices on CH and CRF was also determined.

Methods: Thirty-one myopic subjects were fitted with a daily disposable (Nelfilcon A or Stenofilcon A) in one eye and a monthly disposable (Lotrafilcon B or Comfilcon A) in the other eye. Corneal biomechanical properties (CH and CRF) were measured with Ocular Response Analyzer and corneal topographic indices (anterior surface curvature (simK), eccentricity, surface asymmetry index (SAI) and surface regularity index (SRI)) were measured with the corneal topographer Medmont E 300 before and after three months of CL wear. Changes in corneal biomechanical properties and it's relation with corneal topographic indices, CL materials and lens wear modality were determined.

Results: Sixty-two eyes of 31 subjects with mean age 23.6±3.3 years were included. Twenty of them (64.5%) were females. There was no change in CH ($p = 0.799$) but there was a reduction in CRF by 0.71±1.00 mmHg ($p < 0.001$). Reduction in CRF was correlated with the baseline CRF ($r = 0.598$, $p < 0.001$) but not with power of lens and number of wearing hours per day ($p > 0.05$). It was not associated with lens materials, lens wear modality and gender of the subjects ($p > 0.05$). Although corneal curvature flattened by 0.004±0.07mm, SAI and SRI decreased by 0±0.07, 0.03±0.40 and 0.03±0.33 respectively; all of these changes were statistically non-significant ($p > 0.05$). Change in CRF was correlated with change in SRI ($r = 0.297$, $p = 0.021$) but it was not correlated with change in curvature, SAI and eccentricity ($p > 0.05$).

Conclusion: Three months soft CL wear reduces CRF but it does not change CH. The change in CRF was not associated with the lens materials and lens wear modality. It also was not correlated with changes in corneal topographic indices except a weak correlation with change in SRI.

Key words: Corneal hysteresis; corneal resistance factor; soft contact lens; silicone hydrogel lens; corneal eccentricity; surface regularity index, surface asymmetry index.

Effect of soft contact lens wear on tear film and subjective comfort [CIOCV 2015]

Purpose: The purpose of this study was to determine the effect of three months of soft contact lens (CL) wear on Schirmer score and tear break-up time (TBUT). It was also evaluated the effect of change in tear level on subjective comfort level.

Methods: This was a longitudinal prospective study conducted in normal myopic subjects who had never worn CL before. Schirmer I test and fluorescein TBUT were measured in each subject. Subjects were fitted with a daily disposable (Nelfilcon A or Stenofilcon A) in one eye and a monthly disposable (Lotrafilcon B or Comfilcon A) in the other eye. Level of comfort was subjectively evaluated with a 100 division scale two times a day (after beginning lens wear and before taking out the lens) every day during three months. Average values were used in the analysis. Level of comfort on the first month was compared with the comfort on the third month. Tear level tests were repeated after three months of lens wear.

Results: Schirmer score reduced by 5.8±9.3 mm ($p = 0.000$) and TBUT reduced by 2.8±7.9 secs ($p = 0.009$) after three months of CL wear and these changes were not associated with lens materials (One way ANOVA, $p > 0.05$). Subjective comfort level during the first month and third month remained the same ($p = 0.272$). There was no correlation of tear Schirmer score and TBUT with subjective comfort level ($p > 0.05$).

Conclusion: Three months of soft contact lens wear reduced tear film quantity and quality, however, it did not reduce the subjective comfort level.

Key words: Schirmer score; tear break-up time; subjective comfort level; soft contact lens.

APPENDIX II

DATA COLLECTION FORM

Clinical experimental trials on changes in ocular surface induced by contact lens wear

Sheet No.:_____

Name:_____ **Course:** _____ **Date/time:** _____ ___:___

Profession: _____**Age/Sex:** _____ **Contact Number:** _____

Others:_____ _____

Medical history

General history: _____

Ocular history: _____

Past history: ----------

Allergy: _____

Family history: _____

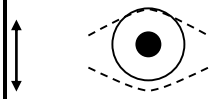
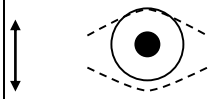
Systemic history: _____

Ocular medication:

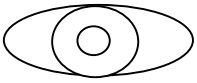
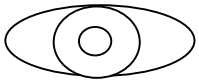
_____

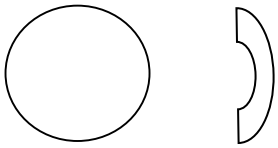
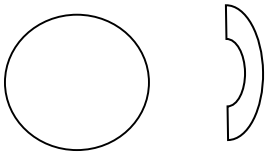
REFRACTION		VA (s/c) OD=	OS=
	CORNEAL TOPOGRAPHY	**OBJECTIVE REFRACTION**	**SUBJECTIVE REFRACTION**
OD	_____(_____) x ____º//____(____) x ____º *(e=____)*	_____ Sph _____ Cyl x_____º	___ Sph ____ Cyl x_____º VA=
OS	_____(___) x ____º//____(_____) x ____º *(e=____)*	_____ Sph _____ Cyl x_____º	__ Sph____ Cyl x_____º VA=

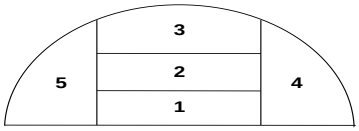
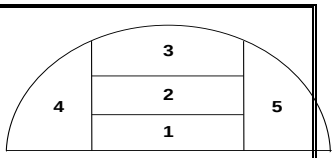
PRELIMINARY EXAMINATION

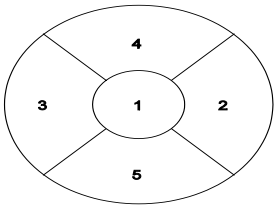
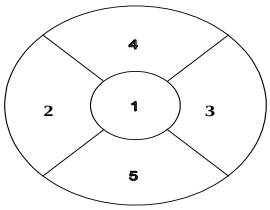
OCULAR PARAMETERS	***OD***	***OS***	**TEAR FILM**	*OD*	*OS*
HVID / VVID	___(mm)/___(mm)	___(mm) /___(mm)	Schirmer test	___(mm/5'')	____(mm/5'')
Pupil diameter (fot./esc.)	___(mm)/___(mm)	___(mm) / ___(mm)	Miniscus height	_____(mm)	_____(mm)
LID POSITION EYE LID TENSION BLINK	 **High∀** **Normal∀** **Low ∀** Com∀ Incomp∀	 **High∀ Normal∀** **Low∀** Com∀ Incomp∀	NIBUT BUT **Others**	**__sg __sg__sg** **__sg __sg__sg**	**__sg __sg__sg** **__sg __sg__sg**
ADITIONAL TESTS	***OD***		***OS***		
Tonometry	**_____mm Hg Time:**		**_____mm Hg Time:**		
Pachometry					
Aberration					
Contrast sensitivity	**RE: A B C D**		**LE: A B C D**		

Goblet cells counts: RE: **A ..... B ..... C .....** LE: **A ..... B ..... C .....**

Ocular investigation	**RIGHT EYE**	**LEFT EYE**
Changes in eye lid and conjunctiva (chalazion, pterygium, pinguecula...)		
HIPEREMIA: BULBAR CONJUNTIVA (0-4) Nasal / Temporal Superior / Inferior	_____ / _____ _____ / _____	_____ / _____ _____ / _____
HIPEREMIA: LIMBAL CONJUNTIVA (0-4) Nasal / Temporal Superior / Inferior	_____ / _____ _____ / _____	_____ / _____ _____ / _____
NEOVASCULARIZATION (0-4) Nasal / Temporal Superior / Inferior	_____ / _____ _____ / _____	_____ / _____ _____ / _____

CORNEAL EPITHELIUM (indicated in number) Microcystes _____/_____ Vacuoles _____/_____ Infiltration _____/_____ Others _____ ___	Front View Section en view 	Front View Section view 
CORNEAL STROMA Striaes, Folds, Others (degree and orintation)		
ENDOTHELIUM POLYMETHETHISM/BLEBS	**Center: ___/___ Peri.:___/___**	**Center: ___/___** **Peri.:___/____**

PALPEBRAL CONJUNTIVA										
	1	2	3	4	5	1	2	3	4	5
Hyiperemia (0-4)	____ S/N	____ S/N	____ S/N	____ S/N	____ S/N	____ S/N	____ S/N	____ S/N	____ S/N	____ S/N
Folicules	S/N	S/N	S/N	S/N	S/N	S/N	S/N	S/N	S/N	S/N
Papillas	____	____	____	____	____	____	____	____	____	____
Others__ _____	____	____	____	____	____	____	____	____	____	____
Irregularities (0-4)										

CORNEAL STAINING										
	AREA 1	AREA 2	AREA 3	AREA 4	AREA 5	AREA 1	AREA 2	AREA 3	AREA 4	AREA 5
Extension (0-4)	_____	_____	_____	_____	_____	_____	_____	_____	_____	_____
Depth (0-4)	_____	_____	_____	_____	_____	_____	_____	_____	_____	_____
Type (0-4)										
Acceptability										
CONJ. STAINING	Nas Temp Sup Inf					Nas Temp Sup Inf				
MGD										
Blephritis										
Others:	SLK Corneal oedema Corneal distortion					SLK Corneal oedema Corneal distortion				

Evaluation sheet

1st trial **Date: ___/___/___**

Time: _____:_____

Lens		Power	Base curve	Diameter	Over Rx	VA
OD						
OS						

Examination time: _____

Comfort	1	2	3	4	5	LD(O)	LI(X)	Centration	1	2	3	4	5	LD(O)	LI(X)
Movement	1	2	3	4	5	LD(O)	LI(X)	Coverage	1	2	3	4	5	LD(O)	LI(X)

Biomicroscopy			
OD			
OS			

Observations:

2nd Trial **Date: ___/___/___**

Time: _____:_____

Lens		Power	Base curve	Diameter	Over Rx	VA
OD						
OS						

Examination time: _____

Comfort	1	2	3	4	5	LD(O)	LI(X)	Centration	1	2	3	4	5	LD(O)	LI(X)
Movement	1	2	3	4	5	LD(O)	LI(X)	Coverage	1	2	3	4	5	LD(O)	LI(X)

Biomicroscopy			
OD			

OS			

Observations:

3rd Trial **Date: ___/___/___**

Time: _____:_____

Lens	**Power**	**Base curve**	**Diameter**	**Over Rx**	**VA**
OD					
OS					

Examination time: _____

Comfort	1	2	3	4	5	LD(O)	LI(X)	Centration	1	2	3	4	5	LD(O)	LI(X)
Movement	1	2	3	4	5	LD(O)	LI(X)	Coverage	1	2	3	4	5	LD(O)	LI(X)

Biomicroscopy

OD			
OS			

Observations:

Follow up sheet	**RIGHT EYE**	**LEFT EYE**

Symptoms *Dryness ∀ Burning ∀ Itching ∀ Photophobia ∀ Unstable vision ∀ Sensaç.Enevoamentoo ∀ Moléstias after __ hours* [LD(O)/ LI(X)]

Others:

Vision ***(on wake up, during day, end of the day)***		
Comfort ***(on wake up, during day, end of the day)***		
Discomfort ***(how often, 0-4)***		
Discomfort ***(how intense, 0-5)***		
Dryness ***(how often, 0-4)***		
Dryness ***(how intense, 0-5)***		
Watery eyes ***(how often)***		

Mantenance *Demasiado Complicado ∀ Realiza Regularmente ∀ Adequadamente ∀ Reacção de Sensibilização ∀ Considera Efectivo o Sistema ∀*

Tempo de porte médio diário: _____

*Hoje:*_____

Eye appearance	1	2	3	4	5	OD	OE)	Centration	1	2	3	4	5	LD(O)	LI(X)		
Movement	1	2	3	4	5	LD(O)	LI(X)	Coverage	1	2	3	4	5	LD(O)	LI(X)		

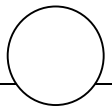
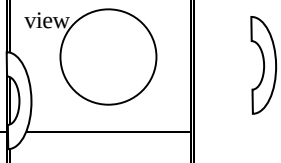
Lens states		

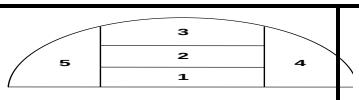
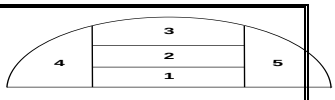
Observations:

VA	OD:	OS:
Over Rx ***(OBJ//SUBJ)***	_____Sph_____Cyl___º // _____Sph_____Cyl___º	_____Sph_____Cyl___º // _____Sph_____Cyl___º
VA c/ Over Rx	OD:	OS:
Keratometry	__,___mm(__,__D)x ___º//__,___mm(__,__D)x ___º	__,___mm(___D)x ___º//__,___mm(__,__D) x ___º

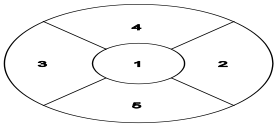
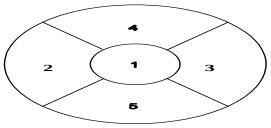
BULBER CONJUNCTIVAL HYPEREMIA (0-4) Nasal / Temporal / Superior / Inferior	_____ / _____ / _____ / _____	_____ / _____ / _____ / _____
LIMBAL CONJUNTIVAL HYPEREMIA (0-4) Nasal / Temporal / Superior / Inferior	_____ / _____ / _____ / _____	_____ / _____ / _____ / _____
NEOVASCULARIZATION (mm) Nasal / Temporal / Superior / Inferior	_____ / _____ / _____ / _____	_____ / _____ / _____ / _____

TEAR FILM NIBUT:__	__ __ :___ __ ___	__ __ __ ___ ___ __
BUT:		
Schirmer test		

CORNEAL EPITHELIUM (indicate in number) Microcysts Vacuoles Infiltration	Frontal sectional view 	Frontal Sectional view 
CORNEAL STROMA Stries, Folds, Others (number and orintation)		
ENDOTELIAL POLYMEGATHISM/BLEBS	**Central:** ___/___ **Periphery.:** ___/___	**Central:** ___/___ **Periphery.:** ___/____

TARSAL CONJUNCTIVA										
	1	2	3	4	5	1	2	3	4	5

Hip/Rug Pap/Fol	__/__ Pap/Fol	___/___ Pap/Fol	___/___ Pap/Fol	__/__ Pap/Fol	___/___ Pap/Fol	___/___ Pap/Fol	___/___ Pap/Fol	__/__ Pap/Fol	__/__ Pap/Fol	___/___ Pap/Fol

CORNEAL STAINING										
	1	2	3	4	5	1	2	3	4	5
Ext/Dep/Type (Aceitabilidade geral)	__/__/__ (__)	__/__/__ (__)	__/__/__ (__)	__/__/__ (__)	__/__/__ (__)	__/__/__ (__)	__/__/__ (__)	__/__/__ (__)	__/__/__ (__)	___/___/___ (___)
CONJ. STAIN.	Nas Temp Sup Inf					Nas Temp Sup Inf				

Others: MGD Blephritis SLK Corneal oedema

Corneal irregularity

IOP:

contrast sensitivity: ***R*****E: A B C D**	**LE: A B C D**
Goblet cell count: ***RE:***	***LE:***

Notes:

APPENDIX III

INFORMED CONSENT

CONSENTIMENTO INFORMADO, LIVRE E ESCLARECIDO PARA PARTICIPAÇÃO EM INVESTIGAÇÃO

Por favor, leia com atenção a seguinte informação. Se achar que algo está incorrecto ou que não está claro, não hesite em solicitar mais informações. Se concorda com a proposta que lhe foi feita, queira assinar este documento.

Título do estudo: Clinical experimental trials on changes in ocular surface induced by contact lenses wear.

Enquadramento: O estudo será realizado no âmbito de um trabalho de doutoramento em Ciências a ser desenvolvido no Centro de Física da Universidade do Minho sob a orientação de Doutora Madalena Lira e Doutora Sandra Franco

Explicação do estudo:

Com este trabalho pretende-se avaliar o estado das estruturas oculares que estão relacionados com o uso de lentes de contacto de Silicone-hidrogel e ainda estudar como algumas das propriedades e parâmetros dessas mesmas lentes ficam alterados com o seu uso. Será realizado uma exame para avaliar as estruturas oculares antes e depois do uso das lentes e mediante a utilização de escalas de graduação pretende-se qualificar objetivamente os possíveis sinais clínicos ou complicações que possam surgir. As propriedades das lentes a analisar serão o índice de refração, quantidade de água, transmitância e ângulo de contacto. Estes parâmetros serão também analisados antes das lentes serem usadas para que se possam fazer as respetivas comparações. No entanto, estes procedimentos serão realizados às lentes de contacto depois de utilizadas e não durante a sua utilização.

O estudo irá decorrer em 3 fases diferentes.

1ª Fase- O objetivo desta primeira tarefa é recrutar pessoas para incluir no estudo. Será realizada a avaliação da superfície ocular e história clinica dos voluntários para avaliar sua conformidade para usar lentes.

2ª Fase - Será executado um exame visual completo para todos os sujeitos envolvidos no estudo. Com este exame pretende-se obter os valores iniciais para selecionar a LC mais adequada assim como a solução de limpeza e armazenamento. Os exames a realizar serão descritos mais è frente neste documento. A todos os sujeitos será adaptada a melhor LC.

3ª Fase - Nesta fase será realizada uma avaliação de rotina e acompanhamento para cada sujeito envolvido no estudo.

Exames a realizar:

Todos os testes utilizados já foram devidamente testados e comprovados a nível mundial. Todas as lentes a utilizar são comercializadas em Portugal assim como os respetivos

produtos de limpeza e desinfeção. Não se pretende desenvolver um novo método, ou um novo material, mas sim avaliar a reposta fisiológicas aos diferentes materiais.

Exames de saúde ocular:

Biomicroscopia: durante este exame pretende-se avaliar o estado da superfície ocular, que engloba, a córnea, conjuntiva (bulbar e tarsal) e pálpebras. Será utilizada uma lâmpada de fenda ou biomicroscópio. A maioria dos exames serão não-invasivos.

Para a realização de algumas medições será necessária a aplicação de fluoresceína sódica. A fluoresceína é um corante vital que pode causar lacrimejo.

Oftalmoscopia: durante este exame avalia-se a retina do paciente. O instrumento a utilizar será o oftalmoscópio e por vezes a luz projetada incomoda o paciente. Este incómodo é provocado pelo encandeamento sendo no entanto passageiro.

Acuidade visual e sensibilidade visual ao contraste: pretende-se avaliar a visão tanto quantitativa como qualitativamente dos participantes com métodos não é invasivos. Não se prevê consequências na realização destes procedimentos.

Exame refrativo objetivo e subjetivo: para o exame objetivo será utilizado o retinoscópio que projeta luz no olho e avaliação das sombras refletidas permite avaliar de uma forma objetiva o estado refrativo do sujeito. Seguidamente é realizado o exame subjetivo mediante a utilização do foróptero. O paciente responde às perguntas feitas pelo investigador de forma a se poder encontrar a melhor esfera/ cilindro e eixo do sujeito. Não se preveem reações como consequência da realização destes exames.

Avaliação das aberrações oculares: As aberrações oculares serão medidas com um aberrómetro de Hartmann-Schack disponível para o efeito e é um procedimento não invasivo.

Topografia corneal: É um exame não-invasivo que permite obter informação sobre a topografia da córnea. O aparelho tem um conjunto de círculos concêntricos que são reflectidos na córnea e cuja imagem é gravada e processada para obter a informação. Não estão previstas consequências na realização destes exames.

Propriedades biomecânicas da córnea: É um exame não-invasivo que permite obter informação sobre as propriedades biomecânicas da córnea. Será utilizado o Ocular Response Analyzer. Com este mesmo instrumento será realizada simultaneamente a avaliação da Pressão Intraocular do paciente.

Avaliação da pelicula lacrimal: A pelicula lacrimal será avaliada tanto quantitativa como qualitativamente. No exame quantitativo será utlizado o teste de Schirmer que consiste na medição da quantidade de lágrima absorvida numa tira de papel especificamente desenvolvida para este fim. Durante a realização do teste de Schrimer pode sentir-se algum lacrimejo reflexo, algumas picadelas na zona do tarso onde será colocado o filtro e a sensação de um corpo estranho. Para a avaliação qualitativa serão utilizadas as miras refletidas na córnea

pelo topógrafo (ao mesmo tempo da realização da topografia corneal). Este teste não será invasivo e não se prevê qualquer sintoma ou consequência na aquando da sua realização.

Adaptação das lentes de contacto: posteriormente à realização de todos os exames anteriores será escolhida a melhor lente de contacto e solução de limpeza para cada sujeito. O sujeito será instruído quanto à utilização das lentes: como retirar, como colocar, como limpar etc, assim como todas as regras de higiene fundamentais para a sua correta utilização. Durante a utilização das lentes podem ocorrer alguns sinais e sintomas considerados normais na utilização de LC. Estes podem incluir: desconforto, secura ocular, prurido e lacrimejo.

Para a realização deste estudo será necessário recrutar voluntários. Este recrutamento será feito entre a população da Universidade do Minho e através do envio de um email onde será explicado o estudo e solicitada a participação. Os voluntários não deverão ser usuários de lentes de contacto., poderão ser de ambos os sexos e maiores de 18 anos.

Condições e financiamento:

A participação será de carácter voluntário podendo desistir a qualquer momento, sem que essa decisão tenha qualquer tipo de consequência.

Não haverá qualquer pagamento de deslocações ou outras contrapartidas financeiras.

As lentes de contacto usadas durante o estudo assim como os produtos necessários para a sua manutenção, serão fornecidos gratuitamente pela equipa de investigação.

Confidencialidade e anonimato: …

Será garantida a confidencialidade e uso exclusivo dos dados recolhidos para o presente estudo.

A identificação dos participantes nunca será tornada pública.

Braga, _____ de _____ de 2013

O investigador: Kishor Sapkota

Assinatura: ----------

Contactos Investigador Principal: Madalena Lira

Email: mlira@fisica.uminho.pt

-o-o-o-o-o-o-o-o-o-o-o-o-o-o-o-o-o-o-o-o-o-o-

Declaro ter lido e compreendido este documento, bem como as informações verbais que me foram fornecidas pela/s pessoa/s que acima assina/m./ Foi-me garantida a possibilidade de, em qualquer altura, recusar participar neste estudo sem qualquer tipo de consequências. Desta forma, aceito participar neste estudo e permito a utilização dos dados que de forma voluntária forneço, confiando em que apenas serão utilizados para esta investigação e nas garantias de confidencialidade e anonimato que me são dadas pelo/a investigador/a.

Nome: … … … … … … … ….... … … … ….... … … … … … … … … … … … … …

Assinatura: … … … … … … … ….... … … … … ... … … … … … Data: …… /…… /……..

ESTE DOCUMENTO **É COMPOSTO POR … PÁGINAS E FEITO EM DUPLICADO: UMA VIA PARA O/A INVESTIGADOR/A, OUTRA PARA A PESSOA QUE CONSENTE**

****The end ****